



# JOURNAL OF AGRICULTURAL RESEARCH

VOL. XIX

WASHINGTON, D. C., JULY 1, 1920

No. 7

## PRACTICAL UNIVERSALITY OF FIELD HETEROGENEITY AS A FACTOR INFLUENCING PLOT YIELDS

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### INTRODUCTION

With the development of a more intensive agriculture there must be a wider use and a progressive refinement of the method of plot tests in agronomic experimentation. Betterment of the method of plot tests must be sought along two lines, (1) the perfection of biological technic and (2) the more extensive use of the modern higher statistical methods in the analysis of the results.

In 1918 Mr. C. S. Scofield, in charge of the Office of Western Irrigation Agriculture, and Prof. E. C. Chilcott, in charge of the Office of Dry-Land Agriculture, asked the writer to undertake an investigation of the statistical phases of the problem of the accuracy of plot tests. The present paper deals with one aspect only of the general problem, that of the lack of uniformity of the experimental field. This is both the most potent cause of variation in plot yields and the chief difficulty in their interpretation.

Many of the careful writers on field experimentation have noted the existence of soil heterogeneity. Few have, however, sufficiently recognized and none have adequately emphasized the importance of this factor.

The problem of field heterogeneity is twofold. First, some measure of the amount of its influence upon crop yields must be obtained. Second, some means of avoiding or of correcting for its influence must, if possible, be secured.

An exact measure of the influence of field heterogeneity, and not merely a vague notion that it may influence experimental results, is the first and most fundamental step in the closer analysis of the factors determining the variability of plot yields. If the application of such a criterion to results obtained by practised agriculturalists from fields selected for their uniformity shows no evidence of heterogeneity, plot tests may be carried out along conventional lines with confidence that

with reasonable precautions reliable results will be obtained. If, on the other hand, the application of such a criterion shows a high degree of irregularity in fields selected for their uniformity by experienced agriculturalists, it is evident that very special precautions must be taken to obtain trustworthy results. Some quantitative measure, and some probable error of this measure, of the amount of irregularity of the soil of a field, as shown by actual capacity for crop production, and not merely a demonstration of its existence is, therefore, required.

The purpose of this paper is to show by the analysis of the actual yields of test plots reported by agricultural experts that the securing of fields suitable for a direct comparison of yields is, practically speaking, an impossibility. The results show that unless special precautions are taken irregularities in the field may have greater influence upon the numerical results of an experiment than the factors in crop production which the investigator is seeking to compare.

The results of this study may seem to be altogether negative—destructive rather than constructive. The unbiased student must, however, admit that a full evaluation of all the sources of error is an essential prerequisite to constructive work. Furthermore, large expenditures of public funds are being devoted to fertilizer tests, variety tests, and rotation experiments. It is preeminently worth while to ascertain to what extent results derived from methods now in use may be considered reliable.

Subsequent papers will treat other phases of the problem.

#### FORMULAE

A criterion of field homogeneity (or heterogeneity) to be of the greatest value should be universally applicable, be comparable from species to species, character to character, or experiment to experiment, and be easy to calculate.

In 1915 the suggestion was made (5)<sup>1</sup> that we may proceed as follows: Suppose a field divided into  $N$  small plots, all sown to the same variety of plants. Let  $p$  be the yield of an individual plot. The variability of  $p$  may be due purely and simply to chance, since the individuals of any variety are variable and the size of the plots is small, or it may be due in part to the diversity of conditions of the soil. If irregularities in the experimental field are so large as to influence the yield of areas larger than single plots,<sup>2</sup> they will tend to bring about a similarity of adjoining plots, some groups tending to yield higher than the average, others lower.

Now let the yields of these units be grouped into  $m$  larger plots,  $C_n$ , each of  $n$  contiguous ultimate units,  $p$ . The correlation between the

<sup>1</sup> Reference is made by number (italic) to "Literature cited," p. 313-314.

<sup>2</sup> Irregularities of soil influencing the plants of only a single small plot may in most work be left out of account, since they are of the kind to which differences between individuals are to a considerable extent due and are common to all the plots of a field.

$\bar{p}$ 's of the same combination plot,  $C_n$ , will furnish a measure (on the scale of 0 to  $\pm 1$ ) of the heterogeneity of the field as expressed in capacity for crop production. If this correlation be sensibly 0 (under conditions such that spurious correlation is not introduced), the irregularities of the field are not so great as to influence in the same direction the yields of neighboring small plots. As heterogeneity becomes greater the correlation will also increase. The value of the coefficient obtained will depend somewhat upon the nature of the characters measured, somewhat upon the species grown, somewhat upon the size of the ultimate and combination plots, and to some degree upon the form of the combination plots.

Knowledge of the values of the correlations to be expected must be obtained empirically.

Let  $S$  indicate summation for all the ultimate or combination plots of the field under consideration, as may be indicated by  $C_n$  or  $\bar{p}$ . Let  $\bar{p}$  be the average yield of the ultimate plots and  $\sigma_p$  their variability, and let  $n$  be constant throughout the  $m$  combination plots. Using the formulae of an earlier memoir (3) in a notation which is as much simplified as possible for the special purposes of this discussion,

$$r_{p_1 p_2} = \frac{\{[S(C_n^2) - S(\bar{p}^2)] / m[n(n-1)]\} - \bar{p}^2}{\sigma_p^2}.$$

This formula assumes the combination plots to be of uniform size—that is, to contain each the same number,  $n$ , of ultimate plots. It may be desirable or necessary to have some of the combination plots smaller than the others.

Such cases are frequently met in practical work. For example, the wheat field of Mercer and Hall is laid out in a 20 by 25 fold manner. This permits only 2 by 5, 4 by 5, or 5 by 5 combinations of the same size throughout. One of Montgomery's experiments with wheat covered an area of 16 by 14 plots which may be combined in only 2 by 2 or 4 by 2 fold groupings to obtain equal areas suitable for calculation. In each of these cases other groupings are desirable.

The formulae are quite applicable to such cases; the arithmetical routine is merely a little longer. The formula is as above, but  $\bar{p}$  and  $\sigma_p$  are obtained by a  $(n-1)$ -fold weighting of the plots,<sup>1</sup> where  $n$  is the variable number of ultimate plots in the combination plot to which any  $\bar{p}$  may be assigned—that is,

$$\bar{p} = S[(n-1)\bar{p}] / S[n(n-1)],$$

$$\sigma_p^2 = \frac{S[(n-1)\bar{p}^2]}{S[n(n-1)]} - \left( \frac{S[(n-1)\bar{p}]}{S[n(n-1)]} \right)^2.$$

<sup>1</sup> That is, each ultimate plot is multiplied by the number less one of the plots in the combination plot to which it is assigned.

Ample illustration of the arithmetical routine has been given in the original paper.

The formulae employed assume the symmetry of the correlation surface. It has been shown elsewhere (4) that spurious values of the correlation coefficient may arise in such cases. Since both  $\bar{p}_1\bar{p}_2$  and  $\sigma_{p_1}\sigma_{p_2}$  take the maximum values when, because of the symmetry of the correlation surfaces,  $\bar{p}_1=\bar{p}_2$ ,  $\sigma_1=\sigma_2$ , it is clear that the limiting value of the spurious correlation will be 0.

Thus it is possible that heterogeneity exists even when  $r_{p_1p_2}=0$ , but a field can not be considered homogeneous if  $r_{p_1p_2}$  has a value which is statistically significant in comparison with its probable error.

Practically, little difficulty will arise from this source, and it can usually be easily avoided by the exercise of a little care in the selection of the proper grouping in doubtful cases.

According to the foregoing conception the relationship between the yield of associated plots is expressed on the universally comparable scale of  $r$ , ranging from 0 to  $\pm 1$ .

When symmetrical tables are used—that is, when each plot is used once as a first and once as a second member of the associated pair— $\bar{p}_1=\bar{p}_2$ ,  $\sigma_{p_1}=\sigma_{p_2}$ , and the regression slope is identical with the correlation coefficient.

Thus, if one ultimate plot,  $p_1$ , of a combination plot be known, the most probable deviation of another plot will be  $p_2-\bar{p}=(p_1-\bar{p})r$ .

Concretely, if the yield of a first plot of a combination plot be 10 pounds above the average of the field as a whole and if the interplot correlation be  $r_{p_1p_2}=0.60$ , the most probable yield of a second plot will be 6 pounds above the average.

Similar reasoning applies throughout. Those who have difficulty in thinking in terms of correlation coefficients can most easily grasp the significance of the results by remembering that in this case the correlation coefficients multiplied by 100 gives the most probable percentage of deviation of the yield of an associated plot when the deviation of one plot of the group from the general average is known.

#### INFLUENCE OF SOIL HETEROGENEITY ON YIELD OF FIELD CROPS

In the paper in which these formulae were suggested it was shown that yield of straw and grain and the nitrogen content of wheat, yield of roots and tops of mangolds, and yield of timothy hay are markedly influenced by irregularities in the carefully selected fields upon which plot cultures have been carried out by agriculturalists.

We have now to ascertain whether this is a general phenomenon or whether it is merely a chance result of these particular cultures. The suggestion has been made that the latter is the case, that with the exercise of a little care uniform fields may be secured, and that substratum

heterogeneity was overemphasized as a factor influencing plot tests. This question can be answered only by actually determining the degree of heterogeneity existing in the fields which have passed the criticism of agricultural experts.

It will be conducive to brevity to have a definite system by which the arrangement of the plots in a field may be described. We shall consider the plots arranged as soldiers in ranks and files. The worker inspects the plot records of a field as recorded on a map or table. By ranks we understand the horizontal rows of plots, by files the vertical rows.

1.80	1.83	2.00	1.91	1.90	1.89	1.79	1.75	2.03	1.83	2.18	1.93	1.77	1.86
1.80	2.07	1.77	1.90	1.70	1.79	1.90	2.04	1.95	1.83	2.06	1.76	1.86	1.79
1.93	1.96	1.83	1.92	1.69	1.90	1.80	1.89	1.83	1.85	2.00	2.13	1.82	1.83
1.89	1.96	1.92	1.86	1.79	1.86	1.79	1.94	1.92	1.80	1.97	2.00	1.87	1.73
2.00	2.01	1.89	1.77	1.97	1.85	1.97	2.10	1.99	1.83	2.00	1.92	1.79	1.89
1.96	1.96	2.00	1.82	1.93	1.82	1.87	1.87	1.92	1.99	1.87	1.83	1.92	1.96
1.89	2.11	1.99	1.87	1.86	1.84	2.06	1.90	1.90	1.82	1.81	1.97	1.79	1.89
2.03	1.86	1.80	1.86	2.06	1.72	1.86	1.72	2.07	1.82	1.84	1.97	1.96	2.01
1.83	1.82	1.82	1.75	1.77	1.72	1.90	1.83	1.90	1.83	1.90	1.85	1.76	2.07
1.87	2.14	1.96	1.87	1.97	1.90	1.90	2.13	1.80	1.83	1.90	2.06	1.94	1.87
1.90	1.94	1.94	1.77	1.89	1.86	1.82	1.87	1.80	1.84	1.87	2.04	1.94	1.89
1.94	1.76	1.96	1.99	1.87	2.04	1.93	1.77	1.74	1.89	1.93	1.96	2.04	1.97
1.83	1.99	1.97	2.08	1.99	1.96	2.15	1.82	1.78	1.83	1.98	1.89	1.85	1.87
1.85	1.87	1.85	1.82	1.92	1.89	2.13	1.82	1.73	1.83	1.96	2.04	1.86	2.08
2.10	1.83	1.85	1.96	2.01	1.92	1.68	1.89	1.85	1.85	1.83	1.85	2.07	1.75
1.93	1.86	1.93	1.87	1.90	1.86	1.99	1.89	1.83	1.82	1.96	1.99	1.99	2.06

FIG. 1.—Montgomery's diagram of 5.5 by 5.5 foot plots of Turkey wheat, showing variations in the percentage of nitrogen in the grain.

Thus figure 1, showing the nitrogen content of wheat plots 5.5 by 5.5 feet given by Montgomery (17), may be considered made up of 16 ranks and 14 files.

In considering rearrangements or combinations of plots we shall refer to the ranks and then to the files—an order easily carried in mind by remembering the trite expression "rank and file." Thus in referring to a 2 by 5 fold combination we mean that two adjacent ranks and five adjacent files of plots were combined. Individual plots may be easily designated. Thus, the plot belonging to the sixth rank<sup>1</sup> and the fifth file in the nitrogen contents of wheat yields contained 1.93 per cent nitrogen.

<sup>1</sup> Ranks are numbered from the top of map, files from the left.

## 1.—MANGOLDS

The yields of 200 plots of mangolds studied by Mercer and Hall (15) may be grouped into combination plots in a 2 by 2 fold manner. When this is done, the correlation between the yields of associated plots has been shown<sup>1</sup> to be as follows:

For weight of roots,  $r = 0.346 \pm 0.042$ ,<sup>2</sup>  $r/E_r = 8.24$ .

For weight of leaves,  $r = .466 \pm .037$ ,  $r/E_r = 12.5$ .

Thus, if one plot of a combination plot is higher or lower than the general average by a given amount, an associated plot may be expected to deviate from the general average by 35 to 40 per cent of this amount.

## 2.—POTATOES

Lyon (14) gives the yield in pounds for each of six sections of a series of 34 rows of potatoes. This crop was harvested from "a piece of apparently uniform land." Each section was 72 feet 7 inches in length. The distance between rows was 34 inches.

Combining yields of rows and of sections of rows by twos, we reduce the field from a 34 by 6 fold to a 17 by 3 fold combination. The correlations between the sections of the rows is then found to be

$$r_{p_1 p_2} = 0.311 \pm 0.043, r/E_r = 7.30.$$

Yield of potatoes in this field is, therefore, markedly influenced by irregularities of soil conditions.

For data on a second test on the influence of field heterogeneity on the yield of potatoes we may avail ourselves of the valuable records of yields of individual hills reported by Stewart (19). Since these are recorded in quadruplets for the purpose of determining the influence of missing hills upon yield,<sup>3</sup> it is not feasible to group them into plots. The influence of heterogeneity may be tested by determining the correlation between the yields of the plants of a quadruplet.<sup>4</sup>

<sup>1</sup> For original data see Mercer and Hall (15, p. 200); also Harris (5, p. 434-435).

<sup>2</sup> The probable errors have in all cases been computed on the basis of the actual, not of the weighted, number of ultimate plots as  $N$ .

<sup>3</sup> The planting scheme adopted was

$$0 \quad a_1 \quad a'_1 \quad b'_1 \quad b_1 \quad 0 \quad a_2 \quad a'_2 \quad b'_2 \quad b_2 \quad 0 \quad a_3 \quad a'_3 \quad b'_3 \quad b_3 \quad \dots$$

where  $a$  and  $a'$  are the two halves of the same tuber and  $b$  and  $b'$  are two halves of another tuber. Thus halves  $a$  and  $b$  were grown adjoining missing hills and were subject to competition on one side only, whereas halves  $a'$  and  $b'$  were subject to competition from two adjacent plants.

<sup>4</sup> Since  $a$  and  $a'$  are halves of the same tuber and  $b$  and  $b'$  are halves of another, the correlations  $raa'$ ,  $rb'b'$  might be due to a specific physiological influence of the characters of the tuber upon both plants developing from the corresponding half tubers rather than to an influence of differences in soil conditions. We have, therefore, determined the correlations between the plants occupying the same relative position in the quadruplet but derived from different parent tubers, that is  $raab$ ,  $ra'b'$ . Hence  $raab$  represents the correlation between the two outside tubers and  $ra'b'$  the correlation between the two inside tubers of the quadruplet. As a control on the results the correlations between one outside and one inside plant have been determined. These are  $raob'$  and  $raob'$ .

The data given by Stewart are number of tubers and total weight of tubers per plant. These two characters permit the determinations of the average weight per tuber.

When all the pairs are omitted which have been omitted by Stewart<sup>1</sup> or have been designated as affected by leafroll, there remain 139 quadruplets. Determining the correlations between the yield of the two plants derived from different tubers but exposed to the same conditions for growth, we have the following correlations:

For number of tubers per hill—

$$\begin{aligned} r_{ab} &= 0.318 \pm 0.051, r/E_r = 6.19. \\ r_{ab'} &= .138 \pm .056, r/E_r = 2.46. \\ r_{a'b} &= .230 \pm .054, r/E_r = 4.26. \\ r_{a'b'} &= .220 \pm .054, r/E_r = 4.04. \end{aligned}$$

For total weight of tubers per hill—

$$\begin{aligned} r_{ab} &= 0.457 \pm 0.045, r/E_r = 10.10. \\ r_{ab'} &= .312 \pm .052, r/E_r = 6.00. \\ r_{a'b} &= .427 \pm .047, r/E_r = 9.09. \\ r_{a'b'} &= .290 \pm .052, r/E_r = 5.53. \end{aligned}$$

For average weight of tubers—

$$\begin{aligned} r_{ab} &= 0.237 \pm 0.054, r/E_r = 4.39. \\ r_{ab'} &= .104 \pm .057, r/E_r = 1.82. \\ r_{a'b} &= .054 \pm .057, r/E_r = .95. \\ r_{a'b'} &= .117 \pm .056, r/E_r = 2.07. \end{aligned}$$

The correlations are positive throughout and generally statistically significant with regard to their probable errors. They show, therefore, that this experimental plot was heterogeneous to an extent that influenced in a very measurable degree the number of tubers, the total weight of tubers, and the average weight of tubers of neighboring hills. For all four measures of interdependence the coefficients are lowest for average weight of tubers and highest for total weight of tubers, while the correlations for number of tubers produced are intermediate in value.

The values of  $r_{ab}$  are consistently higher than those for  $r_{a'b'}$ , notwithstanding the fact that  $a'$  and  $b'$  are more closely associated than  $a$  and  $b$ . The measures of interrelationship between the yields of pairs of plants, one of which occupies an inside and the other an outside position in the quadruplet, are sometimes intermediate between  $r_{ab}$  and  $r_{a'b'}$  and sometimes less than  $r_{a'b'}$ . On the assumption that the correlation is due solely to environmental influence one would expect the highest

<sup>1</sup> Records have been abstracted from Stewart's Table I. Prof. Stewart has kindly furnished some additional information in regard to certain entries in this table.



correlation between the most closely associated plants—that is  $r_{a'b'} > r_{ab}$ . Apparently the reverse condition,  $r_{a'b'} < r_{ab}$ , is due to some

influence of the open space adjoining  $a$  and  $b$ , which allows the fuller development of those plants and in consequence renders them more representative of the extremely localized soil influences to which they are subjected.<sup>1</sup>

### 3.—TIMOTHY HAY

The records of plot yields of timothy hay published by Holtsmark and Larsen (8) have been shown elsewhere (5) to present a correlation between the yield of ultimate plots, combined in a 2 by 2 fold manner, of

$$r = 0.611 + 0.027, r/E_r = 22.4.$$

Clearly the field was highly heterogeneous.

### 4.—ALFALFA HAY

Records of the yields of a series of 46 plots on the Huntley Experiment Farm, Montana, may be used to test further the influence of heterogeneity on the yields of alfalfa hay. Data were kindly placed at my disposal by Mr. C. S. Scofield.

Alfalfa should be of especial interest in the present discussion since it is a deep-rooted perennial herb, whereas all other herbaceous crops investigated have been annuals, or at most biennials.

In field B of this experimental farm there are two series, II and III, each of 23 plots. The 46 plots form a solid block which has been planted each year to one crop just as if it were an ordinary field.

The two series of plots are separated from each other only by a temporary irrigation ditch. Each plot is  $23\frac{1}{2}$  feet wide, 317 feet long, and contains approximately 0.17 acre. These plots have in certain

III		II	
b	a	b	a
230	305	290	305
180	290	240	290
200	310	300	340
210	265	285	355
200	260	300	325
225	285	280	345
215	285	275	365
220	235	270	285
255	235	285	285
210	230	280	260
240	245	300	285
235	235	265	265
230	270	270	295
210	260	270	285
225	260	315	340
225	235	320	330
220	240	275	315
230	200	285	350
255	225	295	340
265	255	310	295
235	225	320	305
250	280	310	315
240	265	310	280

FIG. 2.—Diagram showing yield of alfalfa in first cutting, 1913, on the Huntley experimental tract. The yield is expressed in pounds per half plot.

cases been harvested in subplots of 0.085 acre when the division has been into halves, of 0.0567 acre when the division has been

<sup>1</sup> Possibly competition between closely associated  $a'$  and  $b'$  plants tends to make the yield of one low when that of the other is high.

into thirds, and of 0.0425 acre when the division has been into quarters of plots.

In the spring of 1912 the whole field was uniformly seeded to alfalfa; only one crop was harvested, and yields were recorded for the entire

III				II			
b		a		b		a	
70	95	125	135	135	155	135	175
110	75	85	160	145	125	125	165
80	90	125	110	165	155	150	160
100	65	130	130	145	180	145	180
115	95	110	125	135	165	100	140
115	125	135	135	125	185	130	155
110	95	120	115	145	175	100	155
120	90	100	115	140	150	100	180
100	90	80	105	125	150	45	150
95	95	105	120	125	140	60	145
115	80	95	100	120	140	65	110
115	90	90	105	125	145	120	60
110	100	110	130	120	140	110	115
115	85	120	165	130	150	100	130
105	105	100	145	130	150	145	140
150	95	100	95	100	150	110	115
135	115	90	105	95	110	100	130
155	125	120	100	65	130	115	115
145	130	145	95	120	120	100	115
170	135	155	105	95	135	95	115
135	125	155	95	110	120	115	110
140	115	160	120	110	145	115	130
150	100	120	160	85	150	105	85

FIG. 3.—Diagram showing yield of alfalfa in second cutting, 1913, on the Huntley experimental tract. The yield is expressed in pounds per quarter plot.

plots only. In 1913 and 1914 three cuttings were made. The first cutting was harvested in half plots. The second cutting of 1913 and the first and second cuttings of 1914 were harvested in quarter plots. The

third cutting of 1913 was lost because of a heavy wind which mixed the plot yields at harvest time, so that it was impossible to secure

III				II			
b		a		b		a	
85	85	130	120	130	150	140	165
105	100	105	120	135	150	140	185
100	80	105	110	120	150	170	165
105	110	95	130	165	155	150	170
100	100	105	130	120	140	145	185
100	105	100	125	120	175	195	155
90	100	100	120	155	155	115	200
90	100	105	120	85	155	145	170
120	95	90	120	115	140	170	165
85	95	75	110	155	130	105	155
75	95	85	105	85	130	125	240
60	110	90	100	120	140	160	135
75	100	75	140	95	120	120	130
55	100	75	140	120	130	125	165
75	95	85	125	120	130	140	145
85	100	60	115	125	120	140	160
85	105	100	105	120	135	135	150
115	100	65	115	115	140	155	130
115	125	85	125	150	125	140	130
85	135	95	120	135	135	135	135
105	120	105	105	130	140	165	145
100	115	125	135	140	160	170	140
100	115	140	120	135	120	115	120

FIG. 4.—Diagram showing yield of alfalfa in first cutting, 1914, on the Huntley experimental tract. The yield is expressed in pounds per quarter plot.

accurate weights on any of the plots. The third cutting for 1914 was harvested in subplots one-third the size of the original plots.

The actual yield of these subdivisions is indicated in figure 2<sup>1</sup> for the first cutting and figure 3 for the second cutting in 1913 and in figure 4

<sup>1</sup> Diagrams are set in type instead of being drawn to scale.

for the first cutting, figure 5 for the second cutting, and figure 6 for the third cutting in 1914.

III				II			
b		a		b		a	
100	110	135	125	120	145	145	140
80	85	110	120	130	145	175	155
70	110	140	115	170	155	195	170
70	140	115	125	160	190	145	165
85	125	85	125	180	190	155	175
55	125	95	100	190	175	185	185
65	105	115	115	225	155	200	195
65	110	95	110	190	190	180	165
70	105	100	135	140	155	155	165
110	120	60	100	110	120	100	175
100	110	85	125	95	125	70	140
95	120	120	95	75	100	145	105
110	135	125	135	100	75	125	145
130	120	95	150	135	85	90	170
115	115	100	140	115	125	105	170
130	130	80	115	95	110	95	140
135	115	65	110	110	85	90	150
110	115	80	120	120	130	95	180
145	160	75	135	120	125	105	140
140	135	80	125	105	145	155	100
135	135	90	120	115	155	140	125
120	155	110	130	130	130	135	130
90	160	110	115	120	130	120	75

FIG. 5.—Diagram showing yield of alfalfa in second cutting, 1914, on the Huntley experimental tract. The yield is expressed in pounds per quarter plot.

For the yield of alfalfa on quarter plots for the second cutting in 1913 and the first and second cuttings for 1914 and in third plots for the third cutting for 1914 the correlations are

$$1913, \text{ second cutting, } r = 0.182 \pm 0.048, r/E_r = 3.79.$$

1914, first cutting,  $r = 0.432 \pm 0.040$ ,  $r/E_r = 10.7$ .

1914, second cutting,  $r = .449 \pm .040$ ,  $r/E_r = 11.3$ .

1914, third cutting,  $r = .311 \pm .052$ ,  $r/E_r = 5.99$ .

III			II		
x	y	z	x	y	z
230	190	225	160	240	180
220	170	130	220	220	165
215	150	130	200	205	190
175	150	115	205	190	215
175	155	125	205	220	170
155	155	105	175	160	175
190	130	125	160	175	165
155	145	115	170	165	165
170	105	110	160	155	160
140	120	100	150	120	180
155	90	140	95	160	145
125	125	120	125	165	155
210	100	125	145	160	150
175	140	110	180	165	140
155	145	155	180	195	165
140	115	155	165	185	125
150	125	155	170	170	120
115	120	150	170	150	135
160	150	165	150	165	150
140	165	140	150	165	160
155	155	155	165	195	150
150	175	170	175	160	185
185	150	140	90	155	135

FIG. 6.—Diagram showing yield of alfalfa in third cutting, 1914, on the Huntley experimental tract. The yield is expressed in pounds per third plot.

It will be noted that the results are in very close agreement indeed for 1914. The second cutting for 1913 differs significantly from the others, but no explanation can be suggested.

Grouping all yields in two comparable subplots, we find

1913, first cutting,  $r = 0.407 \pm 0.059$ ,  $r/E_r = 6.93$ .

1913, second cutting,  $r = .343 \pm .062$ ,  $r/E_r = 5.52$ .

1914, first cutting,  $r = .602 \pm .045$ ,  $r/E_r = 13.4$ .

1914, second cutting,  $r = .657 \pm .040$ ,  $r/E_r = 16.4$ .

We note that all the correlations are higher for a 2-fold division than for a 4-fold division. The coefficients for the second cutting of 1913 are again lower than the other values.

The foregoing results are based upon weightings of single cuttings only. It is now desirable to determine the correlations for yield of first and second cuttings combined.

If the combined yield be considered in quarter plots as ultimate units in 1914 we find

$$r = 0.517 \pm 0.036, r/E_r = 14.2.$$

Combining to obtain total yield in half plots in both 1913 and 1914, we have the following correlations between the yields of the two half plots:

For 1913,  $r = 0.387 \pm 0.060$ ,  $r/E_r = 6.46$ .

For 1914,  $r = .709 \pm .035$ ,  $r/E_r = 20.2$ .

#### 5.—STRAW AND GRAIN IN WHEAT

The data of the Rothamsted wheat plots,<sup>1</sup> analyzed in an earlier paper (5, p. 436-440, 443-444), show the following correlations when the 500 plots are grouped in 2 by 2 fold manner for the first 22 files and in a 2 by 3 fold manner for the twenty-third to the twenty-fifth file:

For yield of grain,  $r = 0.336 \pm 0.027$ ,  $r/E_r = 12.5$ .

For yield of straw,  $r = .483 \pm .023$ ,  $r/E_r = 20.9$ .

#### 6.—STRAW AND GRAIN IN RAGI, ELEUSINE, CORACANA

Lehmann (12) has given a series of data derived from the yields of grain and straw of ragi cultivated on the dry-land tract of the Experimental Farm at Hebbel, near Bangalore, Mysore State. The plots used were of 1/10-acre area.

The land was previously owned by several raiyats who have naturally treated it somewhat differently in regard to manuring and cultivation. The various pieces used as garden lands are of course in much better condition than those used for ordinary dry crops. This causes considerable temporary differences to exist in some of the plots in addition to probably slight permanent differences. (12, 6th Rpt., p. 2.)

From these conditions one would expect a high degree of heterogeneity in the series of plots. The data permit the testing of the possibility of a decrease in heterogeneity due to uniformity of crop and treatment for three years.

<sup>1</sup> For data see Mercer and Hall (15, p. 119); also Map B of Harris (5).

These data are, furthermore, of particular interest since they consist of the records of yields for three successive years of the same crop on a series of unirrigated plots in a region where crop production is subject to many uncertainties because of inadequate rainfall.

Fortunately for our present purposes the meteorological conditions during the three years covered by this experiment were very different from year to year. The values of the most significant factor, the July to October rainfall, are given in Table I. This shows that the rainfall in 1906 was practically twice as heavy as in either of the other two years.<sup>1</sup>

TABLE I.—*Rainfall at Hebbel, near Bangalore, Mysore State, India*

Month.	1905	1906	1907	Average of 10 years.
	<i>Inches.</i>	<i>Inches.</i>	<i>Inches.</i>	<i>Inches.</i>
July.....	1.77	7.09	4.17	3.04
August.....	6.75	9.98	1.50	4.32
September.....	1.47	5.50	5.66	8.14
October.....	5.76	8.51	.81	5.97
Total.....	15.75	31.08	12.14	21.47

Maps of the fields are given in the sixth annual report for 1904-1905. Further descriptive detail is given in the seventh, eighth, and ninth reports for 1905-1908. The yield of grain and straw in plots of 1/10 acre grown in 1905 is given in the seventh report. The eighth report gives detail of the crop of 1906 but does not contain the yields, which are summarized for the years 1905, 1906, and 1907 in Tables I and II of the ninth report.

Unfortunately the yields of a considerable number of the plots have had to be omitted from maps I and II of Lehmann's report. In combining in a 2 by 2 fold manner it is necessary either to disregard all combination plots in which there are not four ultimate plots or to weight properly in using those containing 2 or 3 plots only. The course followed has been to group the plots by fours and to determine the correlation by the formulae for a variable number of plots when all of the ultimate plots were not planted.

The following table shows the correlation between the yield of grain, of straw, and of grain and straw:

	1905	1906	1907
Grain.....	0.735 ± 0.031	0.138 ± 0.065	0.716 ± 0.032
Straw.....	.424 ± .055	.164 ± .065	.573 ± .045
Total yield..	.415 ± .055	.145 ± .065	.656 ± .040

<sup>1</sup> A discussion of the growth of these crops in relation to the distribution of the rainfall appears in Lehmann's ninth report (*ib. p. 2-7*).

The results are of unusual interest. In 1905 and 1907 the correlation between yields of grain are unusually high, falling only slightly below three-fourths of perfect correlation. The correlations for yields of straw and for both grain and straw are of medium value in those two years. In 1906, however, the correlations for all the characters are of a very low order; and any one of them taken alone might not be considered significant in comparison with its probable error, which has been calculated on the basis of 103 plots, the number actually involved in the calculations.

Apparently the unusual moisture conditions of 1906 tended to obliterate the differences in the field to which the individuality of adjoining plots was due.

That the unusual weather had a profound influence on the yield of the plots is shown by Table II, in which the means, standard deviations, and coefficients of variation for the yield of the individual plots are set forth.<sup>1</sup>

TABLE II.—Means, standard deviations, and coefficients of variation for the yield of ragi at Hebbel, near Bangalore, Mysore State, India

[Yield expressed in pounds per 1/10-acre plot]

Year.	Grain.			Straw.			Total yield.		
	Mean.	Standard deviation.	Coefficient of variation.	Mean.	Standard deviation.	Coefficient of variation.	Mean.	Standard deviation.	Coefficient of variation.
1905.....	192.8	31.5	16.3	360.8	148.8	41.2	553.5	190.3	34.4
1906.....	136.6	47.1	34.5	191.6	82.0	42.8	328.1	127.4	38.8
1907.....	165.0	48.3	29.3	295.4	80.2	27.1	460.4	126.9	27.6

The means show that yield of both grain and straw was much lower in the abnormally wet year than in either of the others. The standard deviations are of course largely influenced by the actual magnitudes of the yields and are, in consequence, difficult of interpretation. The relative variabilities, as measured by the coefficients of variation, are more orderly. They show that for grain, straw, and total yield the variability of the individual plot yields is greater in the wet year.

Thus the influence of the wet season has not been to make the yield of all the plots alike. It has tended to decrease yield and to increase relative variability from plot to plot. But at the same time it has tended to screen certain factors which in drier years have a marked influence on the individuality of the plots.

Further analysis is not desirable without more detailed information concerning the plots. From the information at hand it seems quite

<sup>1</sup> These constants are obtained by weighting in an  $(n-1)$ -fold manner, since this was the method followed in obtaining the constants for the heterogeneity coefficient.



clear that the innate differences in different parts of the field do not in some seasons exert their full influence upon crop yield because of the weight of other factors. The practical conclusion to be drawn from this result is that an experimental field which might be demonstrated to be sensibly uniform for one crop plant or for one season might not prove to be so for another crop or in a different season.

#### 7.—KHERSON OATS

Kiesselbach (10, 11) has given records of yield for 207 1/30-acre plots of Kherson oats. He says:

These plats were planted . . . upon a seemingly uniform field for the purpose of studying variation in plat yield as a source of experimental error. The entire field had been cropped uniformly to silage corn for a period of eight years. It had been plowed each year and was also plowed in preparation for the oats in 1916. The oats were drilled during two successive days in plats 16 rods by 66 inches . . . The plats were separated by a space of 16 inches between outside drill rows. A wide discard border of oats was grown around the outer edge of the field, so that all plats should have a similar exposure.

Love (13) has shown the existence of heterogeneity in this field.

Grouping the entries of Kiesselbach's Table 27 in a 3 by 1 fold manner the heterogeneity coefficient is found to be

$$r = 0.495 \pm 0.035, r/E_r = 14.$$

For data on a second test of the influence of heterogeneity on the yields of experimental plantings of oats we turn to a small experiment by Montgomery (17), who has given the yields of thrashed grain in grams from 100 consecutive rows of Kherson oats (17, p. 35, Table XIII) each 12.5 feet in length.

The plat chosen for this test was quite uniform and the appearance of the plat at harvest was very satisfactory.

Combining by twos, we find for the correlation between adjacent rows

$$r = 0.339 \pm 0.060, r/E_r = 5.65.$$

#### 8.—GRAIN AND NITROGEN CONTENT IN WHEAT

Montgomery (17, p. 37, fig. 10) has given the yield of grain in grams on 224 blocks each 5.5 feet square. Combining in a 2 by 2 fold manner we deduce

$$r = 0.391 \pm 0.038, r/E_r = 10.2.$$

Again, Montgomery (17, p. 21-22, fig. 7) has given the values of nitrogen content from 224 Turkey wheat plots of the same size. These values are quoted in figure 1 of this paper. The correlation between the plots is found to be

$$r = 0.020 \pm 0.045, r/E_r = 0.44.$$

Finally, Montgomery (16) has given data for both yield of grain and nitrogen content on 224 plots of wheat grown at the University of Nebraska in 1911. The plot (77 by 88 feet) had been sown continuously to Turkey winter wheat for three years.

The plat was of about average uniformity and fertility.

When grouped in a 2 by 2 fold manner these plots of wheat have been shown (5, p. 440-441, map C) to give the following correlations:

For yield of grain,  $r = 0.603 \pm 0.029$ ,  $r/E_r = 21$ .

For percentage of nitrogen,  $r = .115 \pm .044$ ,  $r/E_r = 2.59$ .

Yield of grain per plot is clearly influenced by irregularities of the experimental field, notwithstanding the fact that the plots are only 5.5 by 5.5 feet in area. The correlation for percentage of nitrogen is not certainly significant.

#### 9.—HOPS

Stockberger (20) has given a series of yields for 30 rows of hops which he believes to be quite typical of many thousands of acres in the Sacramento Valley in California. The yields of these rows cover the period of 1909 to 1914. Combining the rows by twos and determining the correlation between the yield of the adjacent rows of the 15 pairs for each of the years, we obtain the following constants:

Year.	Correlation.	$r/E_r$ .
1909.....	$0.444 \pm 0.099$	4.50
1910.....	$.695 \pm .064$	10.91
1911.....	$.061 \pm .123$	8.50
1912.....	$.326 \pm .110$	2.97
1913.....	$.606 \pm .078$	7.79
1914.....	$.386 \pm .105$	3.69
Average.....	.419	5.06

Without exception the coefficients are positive in sign. In general they are fairly large and indicate a substantial degree of heterogeneity in this limited area. Probably the heterogeneity would have been shown to be greater had it been possible to work with yields from the sections of the long rows instead of with the rows as a whole.

#### 10.—UNHUSKED RICE

Coombs and Grantham (2) give the yield in gantangs of a series of 54 square plots  $\frac{1}{4}$  by  $\frac{1}{4}$  chain in dimension.

These plots are arranged in 18 ranks and 3 files. They were harvested from a field of standing rice on which—

the crop was extremely regular, as judged before the cutting, and it had not been subjected to any attack of borer or any devastation of rats or birds.

The yields of the original plots are shown in figure 7. These may be combined in a 2 by 1 fold manner to give a correlation of

$$r = 0.344 \pm 0.081, r/E_r = 4.25.$$

These rice yields taken from a field described as "extremely regular" show that as a matter of fact the field is heterogeneous and that this irregularity influences in a measurable degree the yields of the plots.

13.6	12.0	11.4
14.6	14.0	12.2
14.8	14.4	12.0
13.0	12.4	12.8
15.0	12.0	12.0
13.4	13.8	14.0
14.2	12.2	13.0
14.0	12.0	12.8
14.0	12.0	13.4
14.0	14.0	12.4
15.0	14.0	12.6
14.8	14.0	12.4
14.0	14.0	12.0
14.4	13.6	12.4
12.6	13.0	12.0
12.2	14.0	12.8
11.6	12.0	11.8
12.4	14.0	12.4

FIG. 7.—Diagram showing yield of unhusked rice on Coombs and Grantham's 54 plots  $\frac{1}{2}$  by  $\frac{1}{2}$  chain square. The yield is expressed in gantangs per plot.

#### II.—EAR CORN

Smith (18) has published a series of corn yields for three years on plots of  $\frac{1}{10}$  acre. The yields are given in his original paper. He has kindly supplied the map showing the relative positions of these plots, which are arranged thus:

101, 201, . . . , 601  
 102, 202, . . . , 602  
 . . . . .  
 . . . . .  
 . . . . .  
 120, 220, . . . , 620

Combining yields in a 2 by 1 fold manner, we find for the correlation between the yields of adjacent  $1/10$ -acre plots

For 1895,  $r = +0.830 \pm 0.019$ ,  $r/E_r = 43.4$ .

For 1896,  $r = +.815 \pm .021$ ,  $r/E_r = 39.6$ .

For 1897,  $r = +.606 \pm .039$ ,  $r/E_r = 15.5$ .

It is evident that the field was rather highly heterogeneous.

III				II			
b		a		b		a	
133	132	138	142	136	132	148	140
141	141	132	138	145	135	162	156
135	109	125	135	133	116	147	130
132	153	131	131	130	123	155	150
132	137	135	140	137	112	131	129
135	132	135	131	134	126	126	135
131	128	121	125	126	115	122	136
135	125	128	131	121	115	129	137
133	125	125	130	131	124	129	131
137	124	117	131	127	125	129	132
130	117	119	127	132	129	122	141
134	122	115	125	133	123	119	132
129	122	120	132	130	125	136	137
123	118	125	130	124	124	123	136
129	126	134	129	122	126	127	136
134	124	120	121	126	130	132	136
128	125	115	115	122	123	140	135
128	121	110	110	116	115	125	123
127	124	119	107	114	116	110	115
134	112	121	123	122	126	116	125
145	148	133	125	132	127	126	134
149	154	165	160	162	144	137	130
168	169	165	152	158	169	143	108

FIG. 8.—Diagram showing yield of ear corn, 1915, on the Huntley experimental tract. The yield is expressed in pounds per quarter plot.

For a second test of the influence of field heterogeneity on the yield of ear corn we turn to the Huntley data.

III				II			
b		a		b		a	
78	94	104	128	110	121	132	150
73	81	104	118	116	116	140	142
66	77	84	110	113	102	128	138
66	73	80	99	115	113	128	139
77	79	79	103	116	118	126	127
71	73	86	82	100	110	108	132
76	59	86	90	110	117	111	151
94	65	86	100	102	105	118	116
98	75	80	100	111	101	104	118
88	76	74	99	108	92	102	113
91	82	69	80	100	97	101	101
97	87	83	90	103	92	88	96
75	81	80	107	96	78	96	106
67	76	73	117	95	70	90	117
98	85	74	103	98	84	100	116
111	88	76	97	97	92	110	110
108	88	73	84	84	86	104	115
115	97	66	89	100	87	98	123
104	120	86	100	94	94	97	119
110	106	92	99	96	100	83	104
118	110	100	98	114	108	113	120
108	100	105	110	93	99	117	104
108	98	95	100	103	99	114	98

FIG. 9.—Diagram showing yield of ear corn, 1916, on the Huntley experimental tract. The yield is expressed in pounds per quarter plot.

In 1915 and 1916 corn was grown on the Huntley experimental plots, described above, and was harvested in quarter plots. The yields for the two series are shown in figure 8 for 1915 and in figure 9 for 1916. These records are of special interest in view of the fact that these are irrigated

fields, whereas the data provided by Smith are based on corn grown without irrigation.

Retaining the original division into quarter plots, we deduce for the correlation between the subplots

For 1915,  $r = 0.498 \pm 0.037$ ,  $r/E_r = 13.4$ .

For 1916,  $r = .436 \pm .040$ ,  $r/E_r = 10.8$ .

The results for the two years can not, with due regard to their probable errors, be considered to differ significantly. They indicate a degree of heterogeneity in these Huntley plots quite comparable with that of fields planted to various crops by other observers.

If the quarter plots be combined by adjacent twos and the correlation between the half plots be determined, we find

For 1915,  $r = 0.494 \pm 0.053$ ,  $r/E_r = 9.29$ .

For 1916,  $r = .0431 \pm .057$ ,  $r/E_r = 7.53$ .

The measure of heterogeneity has been only slightly lowered by dividing the plots into halves instead of into quarters.

#### INFLUENCE OF SUBSTRATUM HETEROGENEITY ON YIELD OF ORCHARD CROPS

In the preceding illustrations the crops considered have been herbaceous plants which are generally fairly superficial in their relation to the soil and most of which complete their development in one or two seasons. It seems of particular interest to extend the studies, as Batchelor and Reed (*r*) have done, to the yield of large individual plants, such as orchard trees.

For the purpose we employ the splendid series of data of Batchelor and Reed. They say of their various groves (*r*, p. 251):

The fruit plantations herein discussed, to judge by the surface soil, size, and condition of the trees, as well as their apparent fruitfulness, appeal to the observer as uncommonly uniform. All the orchards studied are situated in semiarid regions and are artificially irrigated during the summer months. This fact is believed to be a distinct advantage for the purpose of reducing the variability of one year's yield compared with another, since it insures a fairly uniform water supply for the soil and reduces one of the variants inevitable in nonirrigated localities.

In the case of the Arlington navel oranges grouped in 8-tree plots as the ultimate unit the authors (*r*, p. 264) report a correlation between plots of  $r = 0.533 \pm 0.085$  when the plots are combined by fours.

It has seemed desirable to test the homogeneity of the soil in each of the orchards studied by them. In determining the following coefficients the individual tree has in each case been the ultimate unit.<sup>1</sup>

Consider first the relationship between the yields of adjacent trees of two navel orange groves.

<sup>1</sup>Yields are reported in pounds per tree of ungraded product.

Grouping the yield of the 1,000 trees at Arlington, shown in figure 1 of Batchelor and Reed, in a 2 by 2 fold manner we find

$$r=0.517 \pm 0.016, r/E_r=33.1.$$

A navel orange grove of 495 trees at Antelope Heights, mapped as figure 2 by Batchelor and Reed, when combined in a 3 by 3 fold manner gives

$$r=0.375 \pm 0.026, r/E_r=14.4.$$

Grouping the 240 Valencia orange trees of the grove shown in figure 3 of Batchelor and Reed in a 2 by 2 fold manner, we find for the correlation between yields

$$r=0.306 \pm 0.039, r/E_r=7.75.$$

For the yield in pounds per tree of Eureka lemons as shown in figure 4 of the authors cited, we find for a 2 by 2 fold grouping

$$r=0.448 \pm 0.028, r/E_r=15.8.$$

This last result is of particular interest, since Batchelor and Reed say of this plantation—

This grove presents the most uniform appearance of any under consideration. The land is practically level, and the soil is apparently uniform in texture. The records show a grouping of several low-yielding trees; yet a field observation gives one the impression that the grove as a whole is remarkably uniform.

Notwithstanding this apparent homogeneity there is a heterogeneity coefficient of over 0.4.

Taking the yields of seedling walnuts in pounds per tree as given in figure 5 of Batchelor and Reed and grouping in a 2 by 2 fold manner, we find

$$r=0.232 \pm 0.038, r/E_r=6.09.$$

Finally, if the yields in pounds per tree of the Jonathan apple trees mapped by Batchelor and Reed in their figure 6 be treated in a 2 by 2 fold grouping, the coefficient is

$$r=0.214 \pm 0.043, r/E_r=4.97.$$

Without exception these groves show material values of the heterogeneity coefficients which are statistically significant in comparison with their probable errors throughout.

#### PHYSICAL AND CHEMICAL BASIS OF THE HETEROGENEITY OF EXPERIMENTAL FIELDS

In foregoing sections it has been shown that when tracts of land are judged by their capacity for crop production the yields are such as to indicate that heterogeneity is a practically universal characteristic of the

fields which may be used for fertilizer tests, variety trials, or any other experimental purpose involving plot yields. In the vast majority of cases the heterogeneity is so great as to leave open to question conclusions drawn from experiments not carried out with all biological precautions and interpreted with due regard to probable errors.

While the actual demonstration of differences in crop yields from one portion of the field to another is the result of final importance from the agronomic standpoint, and while it furnishes all but conclusive evidence that this heterogeneity in yield is due to irregularities in the soil itself, it seems desirable to show that such heterogeneity does actually obtain in the physical and chemical properties of the soil which are determining factors in plant growth.

The desirability of determining the extent to which heterogeneity, in the sense to which the term is used here, obtains in the physical and chemical properties of the soil of experimental fields is emphasized by the following sentences from one of the pioneer papers (21) on the variability of soil samples.

A number of papers have appeared dealing with the variation in the weight of the crop produced over different parts of an apparently uniform field. Such variations reflect the variability of the soil, serving simply as a substratum for the growth of plants, but it is evident that the variations between such measurements as those given do not depend upon the soil as the only variable factor.

At the outset we must recognize that many factors may determine differences in yield. Even if one could secure a tract initially uniform in soil and exposure it is not always possible to be sure that it has all been in the same crop in preceding years. Previous cultures may influence tilth and soil composition by organic remains, by infection with disease-producing organisms, or by differences in the demand of various crops for certain of the plant foods.<sup>1</sup> Such sources of heterogeneity are not readily detected by the eye or by physical or chemical analysis. Even if the experimenter secures a field of sensibly uniform texture, chemical composition, and previous cultural treatment, the uniformity may be readily destroyed in planting or tillage. Rain may interrupt the ploughing, thus exposing the soil of the different portions of the field to air and light for different lengths of time and affecting the physical condition very profoundly. Such sources of error are particularly great in the planting of large experiments. Thus the sources of field heterogeneity can never be fully determined in any case, although individual factors may be demonstrated.

To determine whether an experimental field is heterogeneous with respect to physical or chemical factors, actual measurements of these factors should be made over the field and the heterogeneity coefficient applied. As a first illustration we take a series of soil-moisture

<sup>1</sup>These are factors of particular importance in rotation experiments.



determinations uniformly distributed over a plot on a field at the San Antonio Experimental Farm of the Office of Western Irrigation Agriculture.

Hastings (6) has given a condensed account of the soil conditions of the San Antonio region. A map of the experimental farm by Hastings (7, p. 2) shows the location of field C<sub>3</sub> in which this plot of borings was located<sup>1</sup> and gives meteorological conditions prevailing in 1915, the year in which the borings were made.

Mr. C. S. Scofield kindly informs me that field C<sub>3</sub> had been uniformly treated for some time previously and was in apparently uniform condition. It is nearly level but with a gradual slope to the south and east.

The soil has the superficial appearance of uniformity, but we know from experience that the subsoil, which is usually characterized by a high lime content, is in some

1	2	3	4	5	6	7	8	9	10	11	12	13
14	15	16	17	18	19	20	21	22	23	24	25	26
27	28	29	30	31	32	33	34	35	36	37	38	39
40	41	42	43	44	45	46	47	48	49	50	51	52
53	54	55	56	57	58	59	60	61	62	63	64	65
66	67	68	69	70	71	72	73	74	75	76	77	78
79	80	81	82	83	84	85	86	87	88	89	90	91
92	93	94	95	96	97	98	99	100				

FIG. 10.—Diagram showing location of sample areas examined for soil moisture in a field at the San Antonio Experimental Farm.

places much closer to the surface than in others. However, from a general agronomic standpoint, this field would be regarded as extremely uniform, and observation of it during the growing season would tend to confirm this view.

Borings were made 6 feet in depth and were sampled at every foot.<sup>2</sup> Figure 10 shows the form of this field.

In order to reduce the 100 sample areas to 2 by 2 fold combinations we have discarded the right file and a portion of one rank, retaining only those which can be grouped into fours as indicated by the cross lines. The percentages of moisture content of these 100 sample areas appear in Table III.<sup>3</sup>

<sup>1</sup> The northern border of the sampled area is a line 60 feet south of the north line of the field and parallel to it.

<sup>2</sup> The samples were all taken between March 31 and April 9. During this period there was no rain. Between March 15 and April 10 there were only two rains, one on March 17 of 0.1 inch, the other on March 29 of 0.01 inch. Neither of these was sufficient to affect the soil moisture conditions, since in this region a precipitation of less than 0.25 inch scarcely penetrates the surface-soil mulch. Thus moisture changes during the course of the work can hardly influence the results.

<sup>3</sup> The 12 sample areas which were omitted because of impossibility of combining by fours are starred (\*).

TABLE III.—Moisture content of 100 sample areas of a field at the San Antonio Experimental Farm

[Expressed in percentages]

Sample area No.	First foot.	Second foot.	Third foot.	Fourth foot.	Fifth foot.	Sixth foot.
1	20.2	19.1	17.5	13.1	9.7	8.9
2	23.7	21.6	19.8	16.8	15.0	15.9
3	20.9	20.5	19.4	16.1	15.0	15.1
4	21.5	20.3	18.3	16.0	15.4	14.2
5	23.3	22.4	20.6	19.3	16.1	15.8
6	25.0	24.1	19.7	17.6	16.5	15.3
7	22.8	23.0	20.8	17.0	14.8	14.8
8	24.6	24.3	20.7	18.5	15.8	14.8
9	25.6	25.3	25.3	25.5	23.7	18.7
10	22.9	25.8	26.0	26.2	23.5	18.6
11	28.0	30.4	30.6	29.8	26.8	21.5
12	25.2	25.7	24.5	26.8	24.0	21.2
13*	22.1	22.0	20.1	20.1	16.0	14.9
14	20.2	19.7	17.0	14.5	11.4	9.1
15	22.1	21.3	18.1	14.6	13.8	12.7
16	25.1	21.2	20.0	16.3	15.5	14.1
17	21.8	21.0	19.2	16.6	15.1	14.9
18	23.4	22.4	20.0	16.1	15.7	15.4
19	20.5	20.8	19.6	15.6	13.5	12.5
20	24.0	22.0	19.5	15.1	11.5	9.4
21	20.4	20.7	18.7	13.0	8.1	8.2
22	24.3	24.0	21.0	23.7	21.3	15.2
23	21.3	22.2	21.8	21.7	20.5	16.7
24	24.3	25.7	24.0	22.4	18.3	14.9
25	23.6	23.2	24.4	24.5	22.2	18.4

**TABLE III.**—*Moisture content of 100 sample areas of a field at the San Antonio Experimental Farm—Continued*

[Expressed in percentages]

Sample area No.	First foot.	Second foot.	Third foot.	Fourth foot.	Fifth foot.	Sixth foot.
26*	24.2	23.7	23.0	20.7	19.1	17.8
27	21.1	19.7	18.7	14.7	14.5	17.7
28	21.2	19.6	18.4	17.6	15.2	15.0
29	21.2	20.5	19.6	18.9	17.5	17.1
30	22.9	22.0	19.9	17.5	15.0	14.8
31	21.0	20.7	19.6	16.2	14.0	16.4
32	23.4	21.6	19.3	18.6	16.8	15.9
33	22.2	21.8	20.1	16.6	14.0	14.1
34	23.9	22.7	20.4	17.0	14.6	14.2
35	21.6	20.9	19.2	16.8	15.3	16.3
36	21.4	21.6	20.6	20.0	18.4	16.8
37	25.3	25.6	25.6	24.9	22.2	17.9
38	26.7	29.2	27.0	25.9	23.0	19.1
39*	26.2	29.8	30.4	28.6	26.1	21.5
40	21.8	20.0	19.3	15.7	15.7	16.3
41	19.9	19.4	19.0	15.3	14.8	14.9
42	21.6	20.0	18.2	14.1	15.5	15.0
43	19.6	21.7	19.0	14.7	14.2	13.9
44	21.6	21.7	19.4	15.6	15.3	15.4
45	21.6	20.5	19.3	16.3	7.5	14.2
46	22.6	21.3	12.2	16.2	14.4	14.3
47	21.0	22.0	19.3	15.9	14.7	15.0
48	22.0	21.5	19.8	19.8	14.7	15.2
49	22.7	22.1	20.0	19.5	16.2	16.2
50	21.9	23.3	21.0	19.1	16.2	16.5

TABLE III.—Moisture content of 100 sample areas of a field at the San Antonio Experimental Farm—Continued

[Expressed in percentages]

Sample area No.	First foot.	Second foot.	Third foot.	Fourth foot.	Fifth foot.	Sixth foot.
51	20.0	20.3	19.0	17.6	15.7	17.2
52*	29.6	28.4	27.3	22.0	13.2	16.2
53	20.6	19.8	18.5	15.7	15.9	15.6
54	21.2	20.7	18.8	15.1	14.3	14.5
55	19.3	20.0	18.9	16.3	14.1	14.9
56	21.2	20.8	18.9	16.3	14.1	14.9
57	22.1	21.0	19.5	15.7	15.1	15.7
58	22.7	21.6	19.7	18.3	14.7	15.8
59	21.2	21.0	19.7	17.4	15.2	16.4
60	23.2	22.5	20.7	19.1	16.5	16.5
61	19.4	21.3	19.7	17.8	16.9	17.2
62	22.6	21.3	18.6	18.6	15.0	17.1
63	21.3	20.6	19.3	17.5	17.3	17.9
64	21.7	20.1	18.9	15.8	15.5	16.6
65*	22.5	21.0	20.2	16.7	17.0	20.8
66	20.6	21.2	17.9	17.1	15.8	15.0
67	18.9	19.2	18.2	15.0	14.4	15.0
68	23.4	14.5	19.0	17.7	15.5	15.5
69	21.2	20.4	18.8	17.0	14.0	13.9
70	21.4	20.1	18.4	17.0	15.8	16.0
71	21.0	21.1	18.9	15.6	14.5	15.5
72	22.8	21.4	20.0	16.5	15.5	14.3
73	21.9	21.6	20.2	16.2	14.4	17.9
74	22.8	21.8	20.3	18.0	15.5	17.9
75	21.3	22.8	22.1	21.6	17.7	15.1

TABLE III.—*Moisture content of 100 sample areas of a field at the San Antonio Experimental Farm—Continued*

[Expressed in percentages]

Sample area No.	First foot.	Second foot.	Third foot.	Fourth foot.	Fifth foot.	Sixth foot.
76	21.5	22.0	19.7	17.3	17.2	16.9
77	21.4	21.0	19.7	15.8	15.6	18.0
78*	22.4	21.0	19.2	17.0	16.2	15.6
79	18.5	18.8	18.4	17.0	15.2	15.1
80	20.3	19.6	18.5	15.2	15.0	15.8
81	20.3	20.2	18.8	16.5	14.6	15.5
82	21.5	21.7	18.6	15.8	14.8	14.1
83	20.0	20.4	18.7	15.7	16.3	15.4
84	20.3	20.0	18.9	17.5	14.7	14.7
85	22.4	21.8	21.4	17.1	15.9	14.8
86	23.2	22.0	19.6	16.0	15.7	15.0
87*	21.8	21.6	20.8	19.1	17.2	16.5
88*	23.7	21.8	20.2	16.4	16.9	16.4
89*	28.0	21.6	20.2	18.3	17.0	18.5
90*	23.2	21.7	19.1	16.3	16.3	16.6
91*	22.3	22.9	21.7	19.3	18.5	18.6
92	20.2	19.7	18.2	17.4	14.7	15.0
93	19.0	19.3	18.5	16.1	15.4	15.9
94	22.0	20.4	18.3	16.0	14.9	14.0
95	21.5	19.7	18.8	14.9	14.9	14.5
96	20.8	20.3	18.7	16.3	14.4	15.4
97	20.1	19.5	19.1	17.9	15.0	16.3
98	22.6	20.3	19.4	15.3	15.0	15.3
99	20.4	20.3	18.6	16.4	14.6	14.5
100*	22.6	21.6	19.4	17.5	16.3	15.0

To determine whether the distribution of soil moisture in these plots is such that it might bring about a correlation between the yields of adjacent plots due to heterogeneity in regard to this physical factor in

the field we have merely to determine the correlations between the percentages of water content of associated plots. These are

Depth.	Correlation.	r/Er.
First foot.....	0.317 ± 0.065	4.9
Second foot.....	.529 ± .052	10.2
Third foot.....	.542 ± .051	10.7
Fourth foot.....	.704 ± .036	19.4
Fifth foot.....	.607 ± .045	13.4
Sixth foot.....	.484 ± .055	8.8

The correlations are of a very substantial order, ranging from 0.317 to 0.704. Notwithstanding the fact that there are only 88 stations upon which the probable errors are based, the constants may in every case be considered significant in comparison with their probable errors.

Thus, notwithstanding the fact that we are dealing with a field only 150 by less than 264 feet,<sup>1</sup> there is a marked and statistically significant heterogeneity in respect to so important a factor in plant growth as soil moisture at each level in the upper 6 feet of soil.

This result seems of very real importance in its relation to the practical phases of plot-test work. It shows beyond all dispute that at least under soil conditions such as are found at the San Antonio Experimental Farm, substratum heterogeneity may be very great at levels of the soil which are ordinarily left entirely out of account in the selection of fields which are to be used for plot tests but which are not below the extensions of the roots of the deeper-penetrating crops and not too deep to serve as reserves of soil moisture for the higher layers of the soil in the case of crops which draw their water from more superficial levels.

It is of some interest to determine whether the correlations at one level in the field may be looked upon as sensibly higher than those at other levels. We have, therefore, determined the differences between the correlations at the different depths. These are given with their probable errors, and in relation to their probable errors, in Table IV.

In the table the positive signs indicate higher correlations at lower levels. Of the 10 possible comparisons between the correlations of the first 5 feet, all but one show greater heterogeneity at the lower levels. The sixth foot seems to be somewhat more homogeneous than the second to the fifth foot. A number of the differences are apparently significant in comparison with their probable errors. Thus there is apparently a real difference in the amount of heterogeneity of this field at different levels. Heterogeneity is least at the surface and greatest at a depth of 4 feet.

The significance of this result will perhaps be apparent at once. A field might be reasonably uniform for the surface foot of soil and hence

<sup>1</sup> The total length is 264 feet, but this is reduced by discarding the right file.

fairly well suited to the testing of shallow-rooted crops. Below this it might show a higher degree of heterogeneity. Possibly this heterogeneity of lower-lying strata is the explanation of the large correlations obtained for the yields of neighboring trees in groves planted on apparently uniform soil.

TABLE IV.—Differences and criteria of trustworthiness of differences in the correlation of adjacent plots in soil moisture determinations at various levels

Depth.	Second foot.		Third foot.		Fourth foot.		Fifth foot.		Sixth foot.	
	r.	r/Er.	r.	r/Er.	r.	r/Er.	r.	r/Er.	r.	r/Er.
First foot.....	+0.212 ± .083	2.56	+0.226 ± .082	2.74	+0.387 ± .074	5.22	+0.291 ± .079	3.68	+0.167 ± .085	1.97
Second foot.....			+ .013 ± .073	.18	+ .175 ± .063	2.76	+ .078 ± .069	1.14	— .045 ± .076	.60
Third foot.....					+ .161 ± .062	2.58	+ .065 ± .060	.96	— .059 ± .074	.79
Fourth foot.....							— .096 ± .058	1.66	— .220 ± .066	3.34
Fifth foot.....									— .124 ± .071	1.74

We can pursue this question of the relationship between the water content of the plots somewhat further. If the factors which determine the similarity in the moisture contents of the combination plots affect more than a single layer, we should expect a correlation between the contents of the first and second foot, and so on, in the same boring. The possible correlations have been worked out for the first foot and the remaining layers and are as follows:

Depth.	Correlation.	r/Er.
First and second feet.....	+0.748 ± 0.032	23.59
First and third feet.....	+ .669 ± .040	16.84
First and fourth feet.....	+ .648 ± .042	15.53
First and fifth feet.....	+ .578 ± .043	12.66
First and sixth feet.....	+ .353 ± .063	5.62

There is a statistically significant and even high correlation between the water content of successive levels in the same boring.

When we turn to the problem of chemical heterogeneity, we find that while a number of soil chemists have noted the desirability of considering the variability of the soil in taking samples, the available data suitable for testing the degree of heterogeneity of experimental fields are not extensive.

Kaserer's series of determinations (9) is not sufficiently large or properly distributed over the field to make desirable an attempt to measure heterogeneity. Fortunately Waynick and Sharp (22) have given four excellent series, two for nitrogen and two for carbon, derived from two California fields.

Their samples were taken over a total area of a little more than 1.3 acres on two fields of very different character—a silty clay loam at Davis and a blow sand at Oakley.

The fields were both selected for their apparent uniformity, both being nearly level with no change in the soil mass from one part of the field to another great enough to be detected by the usual field methods. Both fields were practically free from vegetation when selected, and before the samplings were made in March, 1918, all extraneous material had been carefully removed.

Altogether they took 80 samples distributed at 30-foot intervals over the entire area. These samples were arranged in an 8 by 10 fold manner. The original data are given in their Tables 3 and 4. Arranging these in the order of the map of the borings given in their figure 1 and combining in a 2 by 2 fold manner, we derive the following heterogeneity coefficients:

For the silty clay loam at Davis—

For carbon,  $r = 0.417 \pm 0.063$ ,  $r/E_r = 6.67$ .

For nitrogen,  $r = .498 \pm .057$ ,  $r/E_r = 8.75$ .

For the blow sand at Oakley—

For carbon,  $r = 0.317 \pm 0.068$ ,  $r/E_r = 4.65$ .

For nitrogen,  $r = .230 \pm .072$ ,  $r/E_r = 3.20$ .

All these values are statistically significant in comparison with their probable errors. Although the total number of samples is rather small, they indicate in each case a distinct heterogeneity for these important constituents of the soil. Apparently the two fields differ in their heterogeneity, the coefficients for both carbon and nitrogen being distinctly lower on the blow sand at Oakley than on the silty clay loam at Davis. The average carbon content at Oakley is only 0.444 as compared with 1.109 at Davis, while the nitrogen at Oakley is 0.033 as compared with 0.101 at Davis. Probably greater heterogeneity would be expected on general physical considerations on the silt loam than on the blow sand.

The analysis may profitably be carried one step farther. If these fields are heterogeneous in respect to the soil constituents here under consideration, one might anticipate a correlation between the carbon and the nitrogen content of the samples distributed over these fields. The results are

For the Davis loam,  $r_{nc} = 0.785 \pm 0.029$ ,  $r/E_r = 27$ .

For the Oakley blow sand,  $r_{nc} = .744 \pm .034$ ,  $r/E_r = 22$ .



Both constants are large. They show that the field is not merely heterogeneous but that portions which are high in nitrogen are high also in carbon and vice versa.

Waynick (21) has given a series of 81 determinations of nitrification in samples of soil drawn from a field on the University of California farm at Davis.

The field had been planted to corn in 1914, to Sudan grass in 1915, and to grain sorghum in 1916. In 1917 it had lain fallow and was without vegetation when the samples were taken October 20.

The particular area chosen was apparently as uniform as one could well find, being level, of uniform texture and color, and free from small local depression of any kind.

These samples were taken on eight radii of a circle 100 feet in diameter. The samples were separated by a radial distance of 5 feet. Disregarding the one central sample, we may group the remainder by twos in order to determine whether there is a correlation between adjacent samples. The coefficients thus obtained will, of course, not be comparable with those deduced for cases in which the yields or soil samples were uniformly distributed over the field. They will, however, serve to indicate whether or not this field is heterogeneous in the sense that differences prevailed sufficiently large to influence the properties of adjacent samples in a manner to make them more similar than pairs of samples taken at random over the field. His samples were drawn in two series—the first from the superficial 6 inches, the second from the deeper-lying level, 6 to 24 inches.

Waynick's Table 1 gives the residual nitrate in soil as sampled. From it we deduce

For the upper 6 inches,  $r = 0.404 \pm 0.063$ ,  $r/E_r = 6.4$ .

For the subsoil,  $r = .596 \pm .049$ ,  $r/E_r = 12.2$ .

Table 2 gives the nitrate produced from the soil's own nitrogen after 28 days' incubation. We deduce

For the upper 6 inches,  $r = 0.065 \pm 0.075$ ,  $r/E_r = 0.86$ .

For the subsoil,  $r = .059 \pm .075$ ,  $r/E_r = .79$ .

Table 3 shows the nitrate produced from 0.2 gm. of ammonium sulphate in 100 gm. of soil. The correlation coefficients are

For the upper 6 inches,  $r = 0.298 \pm 0.069$ ,  $r/E_r = 4.34$ .

For the subsoil,  $r = .351 \pm .066$ ,  $r/E_r = 5.31$ .

Finally, Table 4 shows the nitrate produced from 0.2 gm. of blood in 100 gm. of soil. The results in this case are

For the upper 6 inches,  $r = 0.120 \pm 0.074$ ,  $r/E_r = 1.62$ .

For the subsoil,  $r = .297 \pm .069$ ,  $r/E_r = 4.32$ .

The coefficients show that for both the upper and lower soil layers there is a correlation of about medium value between adjacent samples for the residual nitrate in the soil. These coefficients are unquestionably significant in comparison with their probable errors.

While the coefficients for nitrogen produced from soil nitrogen after incubation are both positive in sign, neither can be considered statistically trustworthy in comparison with its probable error. When nitrogen is added to the soil, in the form of either ammonium sulphate or of blood, the correlations between the nitrogen produced on incubation are larger. All are positive in sign, and three of the four may be reasonably considered statistically significant.

Thus it is clear that this plot, only 100 feet in diameter, shows distinct heterogeneity in residual nitrate and in the amount of nitrification occurring on incubation after the addition of nitrogen.

#### SUMMARY AND CONCLUSIONS

The purpose of this paper, which is one of a series on the statistical phases of the problem of plot tests, is to show the extent to which the heterogeneity of experimental fields may influence plot yields.

By heterogeneity we understand differences in capacity for crop production throughout the field of such a magnitude as to influence in like manner, but not necessarily to like degree, the yield of adjacent small plots. Thus, variability of plot yields does not necessarily indicate the heterogeneity of the fields upon which tests are made but may be due to other factors.

Heterogeneity is measured by a coefficient which shows the degree of correlations between the yields of associated ultimate plots, grouped in combination plots.

This coefficient has been determined for a relatively large series of experimental fields widely distributed throughout the world and planted to a considerable variety of crops, for which a number of different kinds of yields have been measured. The results show that in every field the irregularities of the substratum have been sufficient to influence, and often profoundly, the experimental results.

It might be objected that by chance, or otherwise, the illustrations are not typical of what ordinarily occurs in plot cultures. But the series considered practically exhaust the available data for such purposes. Furthermore the records are in large part drawn from the writings of those who are recognized authorities in agricultural experimentation and who have given their assurance of the suitability of the fields upon which the tests were made.

For example, Mercer and Hall (15) state the purpose of their research to be—

to estimate the variations in the yield of various sized plots of ordinary field crops which had been subjected to no special treatment and appealed to the eye sensibly uniform.

Their mangolds—

looked a uniform and fairly heavy crop for the season and soil,

while in their wheat field—

a very uniform area was selected.

The data of Larsen were drawn from an experiment—

auf einer dem Auge sehr gleichmassig erscheinenden, 3 Jahre alten Timotheegraswiese.

Montgomery's data were secured from a plot of land only 77 by 88 feet in size, which had been sown continuously to Turkey wheat for three years—

and was of about average uniformity and fertility.

Coombs and Grantham selected a field on which—

the crop was extremely regular as judged before the cutting and it had not been subjected to any attack of borer or any devastation of rats or birds.

Lyon's potato field was selected from—

a piece of apparently uniform land.

Mr. C. S. Scofield kindly informs us that the Huntley tract was selected for apparent uniformity and that prior to the calculation of the constants presented in this paper there was no reason, from general observation, to suspect irregularities in the field. Batchelor and Reed have assured me that their orchards are to all appearances uncommonly uniform. Kiesselbach emphasizes the apparent uniformity of his oat field.

Nothing could more emphasize the need of a scientific criterion for substratum homogeneity than the fact that correlations between the yields of adjacent plots ranging from  $r = +0.020$  to  $r = +0.830$  can be deduced from the data of fields which have passed the trained eyes of agricultural experimenters as satisfactorily uniform.

A second phase of this investigation has been to ascertain whether the physical or chemical requisites for plant growth are so distributed over experimental fields that they may be reasonably looked upon as the source of the demonstrated heterogeneity in yield.

The heterogeneity coefficients for percentage of water content for the upper 6 feet on the Experimental Farm of the Office of Western Irrigation Agriculture at San Antonio, Tex., range from  $+0.32$  to  $+0.70$  and are statistically significant for each of the 6 upper feet of soil. Heterogeneity is least at the surface and greatest at a depth of 4 feet. The surface layer of soil might, therefore, be apparently uniform in water content while underlying layers might differ greatly from one part of the field to another. This may be the explanation of the correlation between the yields of adjacent trees in groves planted in an apparently uniform locality.

Analysis of the data of Waynick and Sharp shows that there is a correlation of from  $+0.23$  to  $+0.50$  between adjacent borings for so important soil constituents as nitrogen and carbon. The correlation

between nitrogen content and carbon content of samples from two different soils is of the order  $+ 0.75$ .

It is interesting to note that these coefficients for water content and for chemical composition of the soil are of about the same order as those found for crop yields. While these results do not prove that the heterogeneity of experimental fields in their capacity for crop production is directly due to these and other physical and chemical factors, there can be little doubt that this is actually the case.

The references here made to the existence of significant heterogeneity in fields passed by agricultural experts as satisfactorily uniform must not be interpreted as a criticism of the work of these investigators. There is, indeed, every evidence of care and thoroughness. The result merely illustrates the inadequacy of personal judgment concerning the uniformity in physical characters or in crop-producing capacity of fields under consideration for experimental work.

The demonstration that the fields upon which plot tests have been carried out in the past are practically without exception so heterogeneous as to influence profoundly the yields of the plots emphasizes the necessity for greater care in agronomic technic and more extensive use of the statistical method in the analysis of the data of plot trials if they are to be of value in the solution of agricultural problems.

To other phases of the problem we shall return in subsequent papers.

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## TRANSMISSION OF THE MOSAIC DISEASE OF IRISH POTATOES<sup>1</sup>

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### INTRODUCTION

In a previous publication<sup>2</sup> evidence was presented that mosaic of the Irish potato is a transmissible disease. In view of the fact that a large number of the experiments establishing the transmissibility of this disease were conducted in the greenhouse, it was considered advisable to confirm those results under field conditions. Furthermore, in connection with these experiments in the field additional contributions to our knowledge of mosaic of potatoes were secured. It will be the purpose of the following pages to present these results, which, unless otherwise indicated, have been obtained in northern Maine.

### TUBER TRANSMISSION

#### MODIFICATION OF SEVERITY FROM YEAR TO YEAR

It is well known<sup>2</sup> that mosaic of Irish potatoes (*Solanum tuberosum* L.) is transmitted from one generation of plants to another through the tubers. It has been shown<sup>2</sup> that there may be great variation in the severity of the symptoms shown by the progeny of a given stock, strain, hill, or tuber.

Progeny of hills which appeared healthy during 1918 while growing in plots which contained some mosaic hills and which were situated near all-mosaic plots were grown and observed during the season of 1919. Most were of the Green Mountain, some of the Bliss Triumph, and a few of the Irish Cobbler variety. Each of the various lots contained some mosaic hills, the percentage varying from 12 to 76. Altogether there were over 4,000 hills, of which 1,200, or 30 per cent, were mosaic. In view both of results reported previously<sup>2</sup> and of the abundance of aphids in 1918, it seems that these mosaic hills represent cases of tuber transmission, following aphid transmission occurring so late in the season of 1918 that no symptoms were apparent. The severity of the symptoms

<sup>1</sup> Conducted as one of the cooperative projects between the Office of Cotton, Truck, and Forage Crop Disease Investigations of the Bureau of Plant Industry, United States Department of Agriculture, and the Department of Plant Pathology of the Maine Agricultural Experiment Station.

<sup>2</sup> SCHULTZ, E. S., FOLSOM, DONALD, HILDEBRANDT, F. M., and HAWKINS, LOB A. INVESTIGATIONS ON THE MOSAIC DISEASE OF THE IRISH POTATO. *In Jour. Agr. Research*, v. 17, no. 6, p. 247-273. Pl. A-B, 25-30. 1919. Literature cited, p. 272-273.

shown by the diseased hills of any lot averaged either "slight plus," "medium," or "medium plus," although it was usually "slight" for many hills and often "bad" for some. "Slight" indicates characteristic mottling sufficiently rare to require careful search; "slight plus" means that mottling is readily apparent but is unaccompanied by wrinkling, "medium" represents both conspicuous mottling and some wrinkling, becoming "medium plus" with marked ruffling and more or less dwarfing; "bad" stands for extreme ruffling and dwarfing which may sometimes cause the mottling to be obscured.

Another similar series of small lots was grown in a second plot. In these the percentage of mosaic hills varied from 4 to 63, being 40 per cent for the 800 hills altogether. The severity of the symptoms shown by the diseased hills was about the same as for the lots in the first plot.

In addition to the healthy hill selections described above, stocks were grown from hills that showed mosaic in 1918. These contained 1,100 hills, of which only 5 had not yet shown mottling by July 30. These 5, of which 4 came from one tuber, were not observed later. It is possible that this healthy tuber, supposedly from an Irish Cobbler hill with bad mosaic, was formed by a long rhizome of a neighboring healthy hill, such as is seen occasionally, and was included with the tubers of the mosaic hill in spite of the precautions usually taken. The severity of the symptoms in the mosaic stocks is indicated in Table I.

TABLE I.—Comparison of mosaic stocks in 1918 and 1919

Variety.	Number of hills		Severity of symptoms, 1918.	Severity of symptoms, 1919.	
	1918.	1919.		Variation between hills.	Average.
Green Mountain.....	25	204	Slight.....	Slight to slight plus....	Slight.
Bliss Triumph.....	34	269	.....do.....	Slight to bad.....	Do.
Green Mountain.....	50	400	Medium.....	Slight plus to bad.....	Medium.
Bliss Triumph.....	18	112	Bad.....	Medium plus to bad....	Medium plus.
Green Mountain.....	20	77	.....do.....	.....do.....	Bad.
Irish Cobbler.....	17	65	.....do.....	.....do.....	Do.

Evidently there was, in the stocks described in Table I, a tendency for the disease to change very little in severity as a result of transmission through the tubers from 1918 to 1919.

Two larger plots, one Green Mountain and one Bliss Triumph, were planted with stock from plots entirely mosaic in 1918. While the percentages of mottled plants on July 9 were, respectively, 67 and 89, all plants were mottled by the last of July. Although in magnitude the plants and yield were inferior to those of comparatively healthy lots, the appearance of the plants and of the plot as a whole was no worse than for the same stock during the three previous seasons.

The foregoing results indicate that mosaic in northern Maine does not necessarily change much from year to year in any diseased stock after the first appearance of the effects of infection. The conditions which determine the severity of the initial symptoms are not yet understood.

#### RELATION TO NUMBER OF TUBERS IN A HILL

The tubers from 10 Bliss Triumph hills and 130 Green Mountain hills, healthy in 1918 but grown near to diseased hills, were all planted uncut in hill lots in 1919. In Table II these hill lots are classified according to the number of tubers in a hill, and the percentage of tubers of each class that transmitted the disease is given.

TABLE II.—*Relation of the number of tubers in a hill to mosaic transmission*

Number of tubers per hill.....	2	3	4	5	6	7	8	9	10	11	12
Number of tubers planted.....	14	27	80	90	180	161	136	81	20	33	12
Percentage of tubers mosaic.....	86	60	32	53	38	46	46	41	30	36	41

There is a high percentage for the classes with two or three tubers to a hill, but otherwise no consistent relation obtains between number of tubers and percentage of mosaic. The results are not modified appreciably if the Bliss Triumph hill lots are disregarded. It thus seems that the increase of mosaic could be reduced by the selection of hills according to yield only if the hills with very low yields were discarded.

#### RELATION TO RELATIVE SIZE OF TUBERS

In connection with the problem of control, the question has arisen whether the selection of tubers according to size would have any effect in regard to the increase of mosaic. Consequently each of the 140 hill lots which are considered in the preceding section was planted in the order of decreasing apparent size of the tubers. With regard to mosaic 69 were mixed—that is, with both mosaic and apparently healthy plants in the same hill lot. In Table III the tubers of mixed lots are classified according to their relative rank, No. 1 being the largest. In addition, the percentage of tubers of each class that transmitted the disease is indicated.

TABLE III.—*Relation of the relative size of tubers in a hill to mosaic transmission*

Rank of tuber in size.....	1	2	3	4	5	6	7	8	9	10	11
Number of tubers planted.....	69	69	67	65	54	43	29	14	8	3	1
Percentage of tubers mosaic.....	67	48	42	40	44	42	38	36	38	33	0

The percentage is high for the group of tubers consisting of the largest ones in the hills and tends to decrease, being 48 per cent for No. 2, 45 per cent on the average for No. 3 to 6, and 36 per cent on the average for No. 7 to 10.



Another way in which to interpret the results is to consider all tubers of a hill lot as occupying equal parts of a line and to determine the "center of disease," which is the point on the two sides of which there are equal numbers of diseased and, if also possible, of healthy tubers. This center of disease was found, for the 69 hill lots described above, to be on the average closer to the large-tuber end of the hill-lot line, 44 per cent of the line being between the two. That is, there was a greater tendency to show mosaic as the relative size of the tuber was greater. However, this tendency is not marked enough to make it seem desirable to experiment further by selecting tubers according to absolute weight or size.

Of 357 hill lots planted in another plot, only the 2 to 6 largest tubers of each were planted, in order of decreasing apparent size. On July 22 to 26, 98 of the hill lots were mixed—that is, partly affected with mosaic. The results are similar to those given in Table III, the percentages being 57, 44, 48, and 35, respectively, for groups 1, 2, 3, and 4. The average center of disease is 46 per cent of the distance from the large-tuber end of the hill-lot line. Before this, on July 2 to 14, only 42 hill lots were mixed; and later, on August 22 to 25, a number of hill lots were either dead or too mature to show mosaic distinctly.

#### RELATION TO POSITION OF SEED PIECE IN THE TUBER

On July 29, 1918, 18 tuber units were observed which had been planted with quartered tubers and were mixed. Of the hills from stem-end quarters, 45 per cent were mosaic, while 62 per cent of those from bud-end quarters were diseased. Likewise there were 24 mixed tuber units of six plants each. Of the hills from stem-end sixths, middle-part sixths, and bud-end sixths, mosaic hills constituted, respectively, 43, 54, and 61 per cent. No attempt was made to sterilize the knife used to cut the tubers.

In 1919 each tuber was cut by means of one of several knives used in rotation and kept, when unused, with blades immersed in 4 per cent formaldehyde solution. Observations made June 28 to July 14 disclosed 44 tuber units, out of 1,109 observed, to be mixed. In these, 48 per cent of the plants from stem-end quarters and 51 per cent of those from bud-end quarters were mosaic. This slight difference had become more marked at the time of the next observation on July 22 to 26, when 84 tuber units, out of 1,348 observed, were mixed. At that time 28 per cent of the plants from stem-end quarters were mosaic, while 61 per cent of those from bud-end quarters were diseased. This difference was reduced slightly when it was found on August 22 to 25 that 20 more of the tuber units were mixed. The preponderance of mosaic in bud-end hills is of no value in the problem of control because of the small percentage of tuber units that are mixed. Its cause is not understood.

## CONCLUSIONS REGARDING TUBER TRANSMISSION

Tubers from mosaic hills may be expected to transmit mosaic. In addition, at least part of those from apparently healthy hills growing near diseased plants will transmit the disease; and they tend to do so more when the parent hill contains only two or three tubers, when the relative size of the tuber in the parent hill is greater, and when the seed piece is nearer the bud end. However, hill selection results in discarding the hills with few tubers. The relation of relative size to mosaic transmission is not sufficiently marked or consistent to justify attempting tuber selection for the elimination of mosaic.

## TRANSMISSION BY GRAFTING

## TUBER GRAFTS

Grafting was attempted with a few tubers by bringing into contact the freshly cut surface of half a mosaic tuber and half a tuber from an apparently healthy hill. In 14 cases the nongrafted half of the supposedly healthy tuber remained healthy, and in 3 of these 14 cases the corresponding grafted half produced mosaic shoots. The three cases of apparent transmission were the only ones of the attempted grafts which established organic union. The failure of transmission in the 11 other cases indicates that mere proximity in a hill was not sufficient for transmission. Furthermore, the small number of successful grafts apparently was due to the fact that relatively old tubers were used.

## DISEASED SCIONS UPON HEALTHY STOCKS

Since transmission by grafting had been somewhat effective both in the field with insects uncontrolled and in the greenhouse with insects controlled,<sup>1</sup> the same method was finally used in the field with insects excluded by means of cages. Three tuber units were used, each consisting of three hills. The untreated plants, the first hill of each unit, remained healthy until dug. In each other hill two or three stalks, from 14 to 17 inches high, were cut down and split, mosaic scions inserted, and contact established with the help of cord and adhesive tape. Soon after the dates of grafting, June 28 and July 2, 1919, the scions died because of shading in the cages; but the branches of the stocks made good growth, and by July 28 a branch in each of two grafted hills was mottled. By August 9 a number of shoots in each cage were mottled and were tagged. At the time of harvest, August 26, these were found to belong to the grafted hills. Healthy stalks also came from these hills but were ungrafted, one even coming from the same seed-piece eye as a grafted stalk.

As the Irish Cobbler variety had not been used for this kind of grafting, six mosaic scions were grafted upon uncaged stalks when the latter were

<sup>1</sup> SCHULTZ, E. S., FOLSOM, Donald, HILDEBRANDT, F. M., and HAWKINS, LOU A. *OP. CIT.*

6 inches high, on June 25, 1919. One scion died immediately, and the hill remained entirely healthy. In the other cases branches from the grafted stalks showed mosaic dwarfing with wrinkling and streak necrosis and some slight mottling in the leaves, while the nongrafted ones remained healthy.

#### TRANSMISSION WITH PLANT JUICE

##### STOCKS TREATED IN 1918

Although several methods of artificial inoculation performed in 1918 apparently had no effect,<sup>1</sup> the high percentages of mosaic shown by some of the 1919 progeny of the treated plants indicate that certain methods were effective. Of 76 plants, progeny of control plants treated with water, 24 per cent were diseased, probably because of aphid transmission in 1918; and of 463 plants, progeny of inoculated plants, 38 per cent were diseased, most of them probably because of aphid transmission. Of one lot of 53 hills, 77 per cent were mosaic. Those developed from progeny of plants which in 1918 were inoculated by means of capillary glass tubes inserted into the petioles immediately after these capillary tubes were taken from a similar position on diseased vines. All of another lot of 28 hills were mosaic. These were progeny of plants whose stems were split and partly immersed for several days in the juice expressed by crushing the tubers of mosaic plants. These two methods may be regarded as promising effective transmission if used in more extensive trials.

##### STOCKS TREATED IN 1919

In view of the fact that mosaic of potato was transmitted by transferring juice from diseased plants to the rubbed and crushed leaves of healthy plants first under greenhouse conditions,<sup>1</sup> it was considered advisable to confirm these results with a larger number of plants and under field conditions. Consequently, during the season of 1919 a series of similar inoculation experiments was conducted in field experimental plots, both in the open and under insect cages.

##### INOCULATIONS WITHIN THE SAME VARIETY IN THE OPEN

The first inoculation was made when the plants had reached a height of from 3 to 8 inches. The juice was expressed from the vines in a grinder and was separated at once from the pulp by straining through cheesecloth. At each treatment the undiluted juice was applied to the leaves after they had been bruised with the fingers. At each inoculation the controls were treated with juice from healthy vines before the plants to be treated with juice from mosaic vines were operated upon. One set of

<sup>1</sup> SCHULTZ, E. S., FOLSOM, Donald, HILDEBRANDT, F. M., and HAWKINS, Lon A. *op. cit.*

instruments was used for the controls and another for the virulent juice. In these experiments the Green Mountain, Bliss Triumph, and Irish Cobbler varieties were used. In each case the juice was taken from vines of the same variety.

The plants of the Green Mountain and Bliss Triumph varieties used in this experiment developed from progeny which in 1918 showed from 11 to 15 per cent of mosaic, eliminated in three roguing. In view of the fact that, with the exception of the Irish Cobbler variety, these were planted by using four seed pieces from a tuber, it was possible to inoculate two of the hills in a tuber unit and have two additional hills of the same tuber unit remaining as uninoculated controls. In each tuber unit the plants in the second and third hills were inoculated—that is, a hill from a stem-end quarter and one from a bud-end quarter. In Table IV are given the results of these inoculations.

From Table IV it is apparent that plants not infected in 1918 if treated with juice from healthy vines remained healthy to the end of the season. (See Pl. 49-51.) As indicated, the exceptions to this result, where some tuber units produced plants which became mottled with mosaic after being treated with juice from healthy plants, were due to the fact that such units had become infected in 1918 in the field but did not present any evidence of infection at the time of the first treatment in 1919.

Plants inoculated with juice from mosaic-diseased vines showed the first mosaic mottling upon the newly developed leaves July 14. At this time aphids were just beginning to appear at the rate of a few individuals to a plant, so that those agents of dissemination can be disregarded as a factor in transmission in these open-field inoculations. It will be noted that with virulent juice a certain number of tuber units showed mottling throughout within a few days after the first inoculation, indicating that the tubers had become infected in 1918. In the remaining inoculated hills every hill, with the exception of one of Bliss Triumph, showed distinct mosaic mottling, while the untreated hills of these same units remained healthy to the end of the season.

In addition to the mosaic mottling, distinct spotting and streaking of the leaves, petioles, and stems obtained by July 25, so that at this time some of the lower leaves began to die. Furthermore, a marked rusting and dwarfing of the leaves also became apparent, so that many of the plants appeared like those in the medium plus or bad stage, indicating that in a single season plants may develop an aggravated form of this disease if inoculated properly. (Pl. 52.)

TABLE IV.—*Inoculations with juice from plants of the same variety*

Variety inoculated.	Source of juice.	Date of inoculation.	Number of tuber units.	Noninoculated hills.		Inoculated hills.			
				Total number.	Number mosaic due to infection.	Total number.	Number mosaic due to infection.	Not mosaic due to 1918 infection.	
								Number mosaic due to 1918 infection.	Percentage mosaic due to 1918 inoculation.
Green Mountain	Healthy Green Mountain	June 20 and 27; July 5 and 12	5	10	4	10	4	6	0
Bliss Triumph	Healthy Bliss Triumph	June 20	5	10	3	10	3	7	0
Irish Cobbler	Healthy Irish Cobbler	June 25; July 2 and 12	5	8	0	8	0	8	0
Green Mountain	Mosaic Irish Cobbler	June 20	10	20	8	22	9	12	100
Bliss Triumph	Mosaic Green Mountain	June 20 and 27; July 5 and 12	10	20	8	20	8	12	100
Irish Cobbler	Mosaic Bliss Triumph	June 20	10	20	8	20	8	12	98
	Mosaic Irish Cobbler	June 24, one inoculation only				12	0	12	100

#### Application of Juice from Plants Showing the Bad Stage of Mosaic

In order to determine whether juice taken from plants badly diseased with mosaic and introduced into healthy plants would induce bad mosaic symptoms in the latter, plants of the Green Mountain, Bliss Triumph, and Irish Cobbler varieties were inoculated in the same manner as those mentioned in Table IV. Three applications at weekly intervals were made upon plants of the same variety as that from which the juices were expressed. The height of the vines at the time of the first inoculation varied from 2 to 8 inches. Plants of five Green Mountain hills, three Bliss Triumph hills, and five Irish Cobbler hills were treated. At the same time also two Green Mountain, three Bliss Triumph, and two Irish Cobbler hills were treated with but a single inoculation.

On July 28, 16 days after the first treatment, the first mosaic mottling was noted upon the inoculated Irish Cobbler vines. By August 15 every inoculated plant, regardless of variety, showed distinct mosaic mottling as well as streaking and ruffling of the leaves as in the bad stage of mosaic; and by August 28 most of the leaves on the lower half of the stems were dead. The plants subjected to but a single inoculation showed symptoms similar to those given three successive treatments, indicating that a single treatment may be sufficient to induce the disease (Pl. 56).

#### INOCULATIONS WITHIN THE SAME VARIETY UNDER INSECT CAGES

##### Early Repeated Application

Juice from crushed mosaic plants (not necessarily mottled at the time of the first inoculation but from stock all mosaic in 1918) was applied to the bruised leaves of two hills in each of three caged tuber units on June 13, 20, and 27, and on July 5. As a control, the third hill in each cage was left untreated; also juice from apparently healthy plants was applied to the bruised leaves of two hills of each of three other caged tuber units on the same dates. In all these cases the plants were from 1 to 6 inches high at the first treatment. On July 9 the topmost leaves of the treated hills in the former three units began to show mottling, which was slight to medium by July 15. On July 30 mosaic branches in these units were ragged and were found at digging time, August 26, to belong to the treated hills, which had no healthy stalks. The tuber units upon which the juice from healthy plants was used remained green and healthy until digging time, while those died which became mosaic.

##### Late Application

On July 14 two hills in each of two caged tuber units were treated with juice from mosaic plants in the same manner as those described in the two preceding sections. Before August 20 the upper leaves of the treated hills became mottled and streaked.

## INOCULATIONS FROM ONE VARIETY TO ANOTHER IN THE OPEN

## Early Repeated Application of Juice

In order to determine whether the juice of a mosaic plant of one variety could induce the disease when introduced into a plant of a different variety of potato, intervarietal inoculations were made under open field conditions. The procedure of inoculation practiced in this connection was similar to that followed with the inoculations indicated in Table IV. In this experiment the control plants always were treated before mosaic juice was used, and a separate set of unstruments was employed for each distinct variety and for juice from each source. Green Mountain, Bliss Triumph, and Irish Cobbler varieties were used. These were subjected to four successive treatments at weekly intervals, as indicated in Table V.

The results given in Table V show that mosaic juice from one variety of potato may produce the disease when introduced into the plants of another variety. In these inoculations the effect upon the treated plants was fully as severe as that obtained when juice was introduced into plants of the same variety, as explained in connection with Table IV. In fact, in many cases the inoculated plants behaved like those in the late or bad stage of the disease. (Pl. 53-55.)

From Table V it is apparent that a large percentage of the plants had become infected in 1918. In view of the fact that such tuber units did not show the mosaic mottling at the time of the first inoculation, when the plants varied in height from 2 to 8 inches, it was impossible to restrict inoculation to healthy units. However, in this connection it is interesting to note that the hills infected in 1918 and inoculated in this experiment showed the disease like the plants in the bad stage whenever the uninoculated control hills in the same tuber units showed but slight or medium infection, so that apparently inoculation with juice increased the severity of the infection which had resulted from transmission in the field the previous season.

Since a considerable number of the plants in this experiment apparently had become infected in 1918, the evident objection might be offered that, in the course of the inoculation, infectious juice was carried from diseased to healthy plants of the same variety and thus caused infection. This objection can be eliminated. Inoculations always were commenced at the same end of the plot and row, and hence the respective tuber units were operated upon in the same consecutive order. In all cases, with the exception of Bliss Triumph inoculations with mosaic Irish Cobbler juices, the inoculated hills of the tuber unit treated first in each of the different varieties became diseased while the uninoculated hills of this unit remained healthy during the course of the experiment. Furthermore, a number of mosaic tuber units, apparently infected in 1918, were among the controls, or the units treated with juice from healthy plants.

TABLE V.—*Inoculations with juice from one variety to another*

Variety inoculated.	Source of juice	Date of inoculation.	Noninoculated hills.		Inoculated hills.				
			Total number.	Number mosaic due to 1918 infection.	Total number.	Number mosaic due to 1918 infection.	Total number.	Per cent mosaic due to 1919 inoculation.	
Green Mountain.	Healthy Bliss Triumph	June 20 and 27; July 5 and 12	10	4			6	0	0
Do.	Healthy Irish Cobbler	June 25; July 5 and 12	10	6	10	4	4	0	0
Bliss Triumph.	do.	do.	10	4	10	4	4	0	0
Do.	Healthy Green Mountain.	do.	10	6	10	6	4	0	0
Do.	Healthy Irish Cobbler.	do.	10	0	10	0	12	0	0
Green Mountain.	Healthy Bliss Triumph	June 20 and 27; July 5 and 12	9	0	9	0	12	0	0
Mosaic Bliss Triumph.	do.	do.	9	3	20	3	17	17	64
Mosaic Irish Cobbler.	do.	do.	10	6	10	6	4	0	0
Green Mountain.	Healthy Bliss Triumph	June 20 and 27; July 5 and 12	10	0	10	0	10	0	0
Do.	Mosaic Irish Cobbler	June 25; July 5 and 12	10	6	10	10	10	6	60
Do.	Healthy Green Mountain.	June 20, one inoculation only.	10	0	10	0	4	4	40
Mosaic Irish Cobbler.	do.	do.	12	0	12	0	12	0	100



In no case did any healthy units become infected even though they happened to be treated immediately after a diseased plant had been operated upon. This indicates that infection does not carry very readily from one plant to another by merely rubbing the leaves of one plant and subsequently practicing the same operation upon a neighboring plant.

#### Late Application

On July 12, 1919, six healthy Green Mountain hills representing three different tuber units were inoculated with juice from mosaic Irish Cobbler vines. A second application was made upon these same plants a week later, when the vines were in blossom.

On August 15 distinct mottling was in evidence on the upper leaves of the vines in each of the six treated hills, and by August 22 some of the leaves were dying in spots and streaks as in the bad stage of mosaic.

Inoculations similar to the foregoing were made July 20 upon the vines of four hills in as many separate tuber units of the Irish Cobbler variety with juices from mosaic-dwarf Green Mountain vines. The plants at the time of the first inoculation had just finished blossoming. By August 20 slight mottling was noted upon the upper leaves of the inoculated vines and also slight streaking of the leaves as in bad mosaic stages. The results in these experiments indicate that plants can be inoculated successfully at the time of blossoming and later, as well as earlier in their development. Also, as stated previously in connection with insect transmission, even though mottling may not be in evidence in the season when infection occurs, nevertheless such plants will not fail to show distinct mottling under favorable environmental conditions during the following season.

#### INSECT TRANSMISSION

##### GREENHOUSE EXPERIMENT WITH APHIDS

Green Mountain tubers furnished by C. I. Gilbert were used at Orono with aphids in a greenhouse experiment because they were expected to be disease-free. This stock was used later in two plots. One consisted of 70 tuber units, of which only 1 was diseased early, evidently as the result of infection in 1918. The other, grown and observed in southern Maine by Dr. W. J. Morse, consisted of 1,357 hills, of which less than three-fourths of 1 per cent were mosaic. In the greenhouse experiment 10 tubers were each cut lengthwise with a flamed knife into four sets and planted on March 17. Half the plants from each tuber were inclosed with insect cages, into each of which about 150 individuals of the common green peach aphid, or spinach aphid (*Myzus persicae* Sulz.) from mosaic potato plants were introduced on April 13 to 16, when the plants were from 2 to 9 inches high. To 15 plants aphids were introduced on leaves on a stick thrust into the soil so that they dispersed without contact between the diseased leaves and the treated plant. To 5 plants they

were introduced on terminal-shoot buds in a flask laid upon the soil. The aphids were killed by nicotine fumigation on April 21. All plants appeared healthy when observed by one of the writers on April 21, when from 10 to 25 inches high. Between April 21 and June 2<sup>1</sup> mosaic symptoms appeared on all of the 15 plants to which the aphids were introduced on sticks. Of the 5 plants to which the aphids were introduced in the flask only 1 became mottled, on July 8. When introduced in the flask many aphids had been injured or killed by water condensing on the interior of the flask following transpiration by the bud. Nineteen of the 20 untreated plants remained healthy; 1 showed slight symptoms on July 9. This plant was the only one found on or before April 28 with uncontrolled aphids upon it—possibly from a mosaic plant or a plant treated with virulent aphids. It was again found to be infested on May 19 and 26. In the case of the 15 plants treated with the stick method of introducing aphids, the percentage showing infection and the average length of the period between treatment and the appearance of the symptoms were greater than in the case of plants treated similarly in a previous greenhouse experiment,<sup>2</sup> probably because in the later trial the plants elongated to heights of from 44 to 72 inches and thus offered for a longer period a chance for the initial display of mottling in the young leaves.

## FIELD EXPERIMENTS WITH CAGES

## EFFECT OF THE USE OF CAGES IN 1918

Although the cages for the control of insects in 1918 did not inhibit completely the dispersal of aphids, nevertheless their use materially checked transmission of mosaic. The effect of these cages upon transmission of mosaic is indicated in Table VI.

TABLE VI.—*Effect of cages on transmission of mosaic*

Variety.	Number of hills selected in 1918.	Number of tubers selected for 1919 planting.	Treatment in 1918.	Percentage of mosaic in 1919.
Green Mountain.....	9	32	Uncaged.....	49
Do.....	3	6	Caged with mosaic hill.....	100
Do.....	22	50	Caged.....	0
Bliss Triumph.....	31	66	Uncaged.....	35
Do.....	20	54	Caged.....	0

The number of hills reported in Table VI includes only a small percentage, a representative lot, of the total number planted in 1918. However, each hill indicated was grown under a separate cage. While these

<sup>1</sup> Observations after May 1 were made weekly by Viola L. Morris, laboratory assistant, and finally by Dr. W. J. Morse, neither having any information regarding the previous treatment of any plant.

<sup>2</sup> SCHULTZ, E. S., FOLSOM, Donald, HILDEBRANDT, F. M., and HAWKINS, LOU A. *OP. CIT.*

results might be interpreted as suggesting that some insect besides aphids was a deciding factor, it is possible for the aphids observed in the cages late in the 1918 season to have come from a very few which did not carry mosaic, and as yet no other insect is known to transmit mosaic of potato.

#### EXPERIMENTS WITH APHIDS

Small colonies of the peach aphid were brought from Orono May 1 on radish and mosaic potato plants. Both increased while feeding on these plants. On June 7, when the vines were from 1 to 4 inches high, 9 caged plants of 3 tuber units were treated with aphids from radish plants, about 150 to each hill. These 3 units were regarded as controls, since the aphids had lived for a number of generations on radish plants and were supposed to be free from a mosaic virus. Three caged plants of a fourth tuber unit were treated with aphids from a mosaic potato plant; 2 plants were left in each hill and the aphids, about 100 to a hill, were introduced on leaves on a stick thrust into the soil near each hill. On June 30 and July 5, when aphids were very numerous, these 12 plants were sprayed with a solution of soap and nicotine sulphate. The plants used came from the Gilbert stock, already described as exceptionally healthy. On July 28 the fourth tuber unit was slightly mosaic in some branches of 1 hill, and by August 9 it was dead, as the result of excessive aphid infestation. The 3 controls remained healthy until dug on August 26.

On June 17, nine half-tuber sets from stock caged in 1918 were planted under three cages. On June 28, when the vines were from 1 to 3 inches high, the plants were treated with aphids from mosaic plants; several hundred aphids were introduced by each hill with the stick method described above. They were sprayed on July 5 and 8. On August 9 one hill showed some mosaic. When dug on August 26, this hill was all mosaic, while two other hills—one in the same cage—were each mosaic in the upper leaves of one stalk. The untreated plants from the other nine half tubers were grown in the field and remained healthy throughout the season.

Four tuber units of the Gilbert stock, comprising 12 hills, were treated on July 12, when the plants were large enough to press against the tops of the cages. The first unit was treated with hundreds of aphids from radish plants and the others with aphids from mosaic potato plants. In the latter case several thousand aphids were left on the diseased leaves and stems in a flower-pot saucer set at the base of each of the first and third hills, whence they dispersed within a few days. The first tuber unit remained healthy throughout the season. The other three were still healthy on August 9, but when dug on August 26 two hills were mosaic, each in the upper leaves of one branch of a stalk.

## EXPERIMENT WITH FLEA BEETLES

Three caged tuber units (9 hills) of the Gilbert stock were treated with flea beetles (*Epitrix cucumeris* Harris) on June 13, 1919, when a few inches high. The middle hill of each unit was covered with a cylindrical cage set inside the larger cubical one; the other two hills were treated with several hundred flea beetles. These insects were collected from small potato vines which developed from 100 per cent mosaic stock. On June 20 the cylindrical cages were removed and most of the flea beetles, which had damaged the plants considerably, were driven out of the cages or killed by hand. On June 16 two more similar tuber units were treated likewise. All the hills remained healthy until dug on August 27.

As controls, four similar tuber units were treated in the same way, except that the beetles were taken from plots of mostly healthy potatoes or, in one unit, from bushes near the potato field. All the hills remained healthy until dug on August 26.

## EXPERIMENT WITH COLORADO POTATO BEETLES

Five caged tuber units (15 plants) of the Gilbert stock were treated with Colorado potato beetles (*Lephotarsa decemlineata* Say.) on July 3, 1919, when they reached nearly to the tops of the cages. The insects were gathered with brush and pan from plants in all-mosaic plots when from 2 days old to two-thirds full grown. Two stalks were left in a hill, and the first and third hills in every cage were treated with over 100 of the larvæ each. These were shaken from the gathering pan upon a cloth and were either rolled upon the leaves or left on the cloth while it was laid on the plant. Within 24 hours the plants had been damaged rather severely. They were sprayed with an arsenical poison, which soon caused the death of the larvæ. All the plants remained healthy until dug on August 26 and 27.

Three similar tuber units were treated likewise on July 7, except that the larvæ were obtained from plants in plots almost disease-free. These also remained healthy until dug on August 27.

## FIELD OBSERVATIONS WITHOUT CAGES

## GREENHOUSE STOCKS

Tubers from the 53 plants used in the first aphid experiment performed in the greenhouse at Orono<sup>1</sup> were planted whole. All of the 37 tubers from plants which became mosaic after the introduction of aphids from mosaic potatoes produced diseased hills, except 2 which came from a plant with 3 out of 7 stalks apparently healthy. The 2 healthy tubers were probably produced by the 3 healthy stalks. All of the 10 tubers from plants which remained apparently healthy until harvested, although they were fed upon by aphids from mosaic plants, were

<sup>1</sup>SCHULTZ, H. S., FOLSOM, Donald, HILDEBRANDT, F. M., and HAWKINS, LOU A. OP. CIT. P. 25-30.

mosaic. None of the 38 tubers from caged untreated plants or of the 15 from plants fed upon by aphids from a healthy potato plant were mosaic. Of the 34 tubers from uncaged and untreated plants 1 was mosaic; it came from a half-tuber hill that early showed ruffling and chlorosis along the veins but no typical mosaic mottling such as was shown, in addition to these incomplete symptoms, by the corresponding half-tuber hill after treatment with virulent aphids. Of the 37 tubers from plants fed upon by aphids from radish plants 4, or 11 per cent, were mosaic; these 4 came from 2 plants recorded as having been fumigated to eliminate a few aphids which were found on them and which were of unknown origin, possibly from neighboring diseased plants.

These results agree essentially with those which were secured previously with the first generation of the same stocks and which were described to prove the possibility of transmission by aphids. They also indicate that (1) mosaic mottling may be restricted to the parts of the leaf along the veins, (2) a plant with three stalks healthy and four mosaic may produce three mosaic tubers and two healthy ones, thus explaining the partial infection of hill lots, (3) plants treated with virulent aphids may appear healthy but produce progeny that are all mosaic, as shown previously by the writers,<sup>1</sup> and (4) apparently healthy plants inspected often for aphids and fumigated to eliminate these insects as soon as they are discovered may produce progeny of which a small percentage is mosaic.

In connection with the experiment just considered it was necessary to treat a number of control plants by laying a mosaic leaf upon each. These were kept in a different greenhouse room where aphids were more abundant, and they were never caged. Of 45 tubers from these, and also of 25 tubers from similar plants with no leaf laid on, 20 per cent were mosaic, all coming from plants recorded as being fumigated to eliminate uncontrolled aphids found upon them.

#### PROXIMITY STUDIES WITH PLOTS

In 1918, plots 1, 2, and 3 were each rogued of mosaic hills three times. Stocks from the first two, Green Mountain and Bliss Triumph, respectively, each showed mosaic in 20 per cent of the hills in 1919, while that from No. 3, Green Mountain, next to No. 4, a Green Mountain plot with 45 per cent of the hills diseased, showed mosaic in 30 per cent of the hills. In 1919, each of the stocks was rogued several times and grew between similar stocks. All these plots in both years were each  $\frac{1}{4}$  acre in area. The greater percentage of mosaic in 1919 in stock from plot 3 can be explained best by the greater proximity in 1918 to a half-mosaic plot and by consideration of the apparently greater ease of dispersal of aphids, which were numerous in 1918, from the half-mosaic plot to No. 3. Plots 1, 2, and 3 were planted with stocks A, B, and C, respectively, described in Table VII.

<sup>1</sup> SCHULTZ, E. S., FOLSOM, Donald, HILDEBRANDT, F. M., and HAWKINS, LOO A. *OP. CIT.*

## INTERSEASONAL INCREASE

It was very apparent that aphids, which seemed as abundant as the flakes in an ordinary snowstorm when they were migrating in the late summer, were unusually numerous in 1918. Consequently it is of interest to compare the relative interseasonal increase in mosaic in the same stocks from 1917 to 1918 with that from 1918 to 1919. It has been demonstrated in several greenhouse experiments already discussed that aphids may transmit mosaic without the symptoms being shown until the progeny of the inoculated plants is grown the following season. It is considered that they may do likewise in the field during the latter half of the summer, which is usually the only time when they are abundant on potatoes in northern Maine, although there are more species in the field than were used in the various experiments.

Previous experiments seemed to indicate that the percentage of mosaic in susceptible varieties could be materially reduced by roguing the diseased plants from the plots as soon as the mottling appeared upon the vines. However, before it was fully demonstrated that insects were capable of transmitting mosaic the plots usually were arranged in such a manner that insect transfer could take place very readily. In view of this situation, it is possible to note the effects of those agents of transmission upon the performance of a few of the plots, each including  $\frac{1}{4}$  acre, which were rogued during the last three seasons. Table VII records the observations on these plots.

TABLE VII.—*Relation of aphids to increase of mosaic from season to season*

Variety.	Stock.	1917.			
		Location.	Per-centage of mosaic.	Treatment.	Number of aphids.
Green Mountain...	A	Next to 100 per cent mosaic stock.	32	Rogued three times.	Few.
Bliss Triumph....	B	.....do.....	44	.....do.....	Do.
Green Mountain...	C	.....do.....	49	Rogued once.	Do.

  

Variety.	Stock.	1918.			1919.
		Location.	Per-centage of mosaic.	Treatment.	Per-centage of mosaic.
Green Mountain...	A	Six rows from 45 per cent mosaic stock.	11	Rogued three times.	Very abundant.
Bliss Triumph....	B	Nine rows from 45 per cent mosaic stock.	16	.....do.....	.....do.....
Green Mountain...	C	Next to 45 per cent mosaic stock.	13	.....do.....	.....do.....

It will be noted in Table VII that in 1917 certain factors seemed to be more favorable for the spread of mosaic than in 1918—namely, higher percentage of diseased hills (rogued) in the plots, greater proximity of unrogued mosaic stock, and higher percentage of mosaic in the nearest unrogued diseased plot. However, there was less spread in 1917 than in 1918, as shown by the lower percentage of mosaic in 1918 than in 1919, in correlation with the greater abundance of aphids in 1918. Furthermore, these observations indicate how difficult the problem is of producing perfectly mosaic-free stocks from susceptible varieties wherever these agents of transmission exist.

#### EFFECT OF VARIATION IN THE TIME OF HARVESTING IN 1918

It was expected that if aphids were a deciding factor in mosaic transmission the lots of tubers harvested at progressively later dates during their increase in numbers would show an increasing percentage of mosaic. Seventy-eight healthy hills (66 Green Mountain and the rest Bliss Triumph or Irish Cobbler) were selected in 1918 in a plot containing many small lots all with more or less mosaic. Aphids became noticeable on potatoes the last part of July and increased in numbers so that they were very numerous about the middle of August and more excessively abundant as the end of the month was approached. Tubers about an inch in diameter were harvested on August 8 but did not keep with the methods used. Another set of tubers was harvested on August 15 and a third on August 26, one tuber being removed from every hill on each date. On September 12 the remaining tubers—321 in all—were harvested. The tubers were planted uncut in 1919 and transmitted 6, 14, and 50 per cent of mosaic, respectively, for the three lots. Apparently some of the infection occurred before August 15, but most of it was too late to affect many of the tubers harvested by August 26. This difference can be explained best by the great increase of aphids during August, together with the results obtained in the experiments on aphid transmission.

#### TEST OF THE SEED-CUTTING KNIFE

In 1919 stock was available from 1918 all-mosaic plots and rogued plots. One hundred tubers from the former were divided by three parallel transverse cuts so that no two cut surfaces joined in a seed piece, while 100 tubers from the latter were quartered by a transverse and a longitudinal cut so that each seed piece had two cut surfaces joining at a right angle. The same knife was used, cutting alternately tubers from the two lots. The 800 sets were left mixed in the same sack for over a day and planted by hand at 15-inch intervals in two rows. Another mixture was prepared in the same way with 200 tubers from the same two barrels, but in this case the pieces from the all-mosaic lot were sorted

out and discarded and only the others were planted. The latter occupied the third row, and the fourth row was used for a control lot prepared similarly except that no all-mosaic stock was used. Upon examination of the four rows on July 23 the control row was found to contain 85 mosaic hills, the third row 72, and the first two 473—that is, 75 excluding the 400 from all-mosaic stock. No change in the number of mosaic hills was found on August 18. A  $\frac{1}{4}$ -acre plot of the rogued stock was planted elsewhere and contained 80 mosaic hills in each 400. Evidently the furnishing of conditions apparently optimum for knife transmission had no effect upon the mosaic percentage.

It was thought in 1918 that the partial infection of tuber units might be due to knife transmission. As stated before (p. 318), in 1919 when tuber units were planted three knives were used in rotation, each one being immersed in a 4 per cent formaldehyde solution when not in use. However, the partial infection of tuber units and hill lots was as common as before.

#### TESTS OF EFFECTS OF CONTACT

##### GREENHOUSE EXPERIMENTS

As has been reported,<sup>1</sup> out of nine healthy plants kept in contact with mosaic plants in a greenhouse one showed mosaic, but not until after a few uncontrolled aphids, possibly from mosaic plants, were discovered upon it. At about the same time, March 13, 1919, each of 12 tubers was split into three sets and planted in small pots. The plants from 4 tubers became mottled by April 1 when from 3 to 13 inches tall. The other 24 were transplanted about April 1 into large pots, 2 from each tuber into steam-sterilized soil and the third into soil containing a mosaic plant. The transfer was made by knocking off the bottom of the small pot and setting it into a hole formed by a small empty pot put in when the mosaic set was planted. The method used permitted the mingling of the roots of the two plants while it kept the two sets of tubers mostly apart and facilitated harvesting them separately. The vines of the two plants were twisted and tied together. All of the 24 plants remained healthy until July 9.<sup>2</sup> They had ceased to elongate by this time and soon afterwards were dug. The tubers were not planted, because of the abundance of aphids on the plants in July.

##### FIELD EXPERIMENTS WITH INSECT CAGES

Nine tubers of the Gilbert stock were planted halved in 1919, each two sets being separated by a mosaic set and all three caged. On July 30 three of the mosaic hills were dead or nearly so. The Gilbert hills all remained healthy until August 9 and when dug on August 27

<sup>1</sup> SCHULTZ, E. S., FOLSOM, DONALD, HILDEBRANDT, F. M., and HAWKINS, LOU A. OP. CIT.

<sup>2</sup> Observations after May 1 were made weekly by Viola L. Morris, Laboratory Assistant, and finally by Dr. W. J. Morse.



were entirely healthy except for mosaic mottling in the few uppermost leaves of several branches of a stalk in one hill. These leaves appeared young. They had evidently been pushed hard against the inside of the cage and had a very few aphid skins and aphids clinging to them. They may have been infected as the result of contact before aphids entered the cage, by aphids on the outside of the cloth against which the leaves were pressed, or by aphids that came from mosaic plants in the next row and that entered through a small hole that was found to have been made accidentally in the cloth.

#### FIELD EXPERIMENTS WITHOUT CAGES

As was pointed out in a previous section regarding the test of the seed-cutting knife, the mixing of all-mosaic stock and rogued stock in two rows was not followed by a higher mosaic percentage for the rogued stock than was shown by it in a control row. The negative results in this case do not disprove the possibility of infection occurring too late to be evident during the current season—that is, after the roots and vines have become intertwined.

In 1918 five Green Mountain hill lots were found to be partly mosaic. The healthy hills were harvested separately, were classified according to their proximity in the row to a mosaic hill, and the tubers were planted uncut in 1919. Twenty-eight tubers were progeny of plants each of which grew between two mosaic hills, and 54 per cent of them were mosaic. Eighty-nine were progeny of hills each of which was between a mosaic hill and a healthy hill, and of these 63 per cent were mosaic. On the other hand, 40 per cent of the 220 tubers from hills each of which grew between two healthy hills were diseased. If these 220 tubers are arranged in five groups, H<sub>1</sub>, H<sub>2</sub>, H<sub>3</sub>, H<sub>4</sub>, and H<sub>5</sub>, according to the increasing number of healthy hills between the parent and the nearest mosaic plant in the row, the groups contained, respectively, 75, 53, 41, 33, and 18 tubers, with 56, 24, 54, 24, and 17 per cent of them diseased. Since being next to a mosaic plant in the same row seemed to increase the chance of infection as much as 54 or 63 per cent is greater than 40 per cent, it evidently is a contributing factor in mosaic transmission; but judging from the varying percentages of infection among the classes of plants which were not next to mosaic hills in the same row, it probably aids in the spread of the disease only by aiding aphid transmission.

A slightly different type of experiment consisted in comparing the progeny of three small 1-row Green Mountain lots, of from 100 to 200 hills each, from which the mosaic hills (respectively, 6, 16, and 30 per cent) were removed on August 1, 1918, with two similar lots from which the mosaic hills (respectively, 6 and 18 per cent), together with each healthy hill next in the row to a mosaic one, were removed

August 1. In spite of the differences in contact with diseased hills, the progeny of the two lots were 27 and 35 per cent mosaic, respectively, and the progeny of the three lots were from 25 to 35 per cent mosaic. Aphid dispersal from neighboring mosaic plots was easy, and it apparently nullified any effect that the difference in contact might have had.

#### TEST OF SOIL HARBORING

##### GREENHOUSE EXPERIMENT

At harvesting time in 1918 one tuber was taken from each healthy hill in two hill-selected lots. At Orono on January 14, 1919, these tubers were split with a flamed knife, and one set was planted in steam-sterilized soil and the other in soil from which a mosaic plant had been removed on December 30 or January 13. Nineteen pairs of half tubers were used, and the plants from 7 pairs were mosaic by February 22, when from 1 to 20 inches high. The plants from the other 12 pairs reached their maximum height about March 5 and remained healthy until dug in April. The second generation of the 12 healthy pairs was grown and found to be entirely free from mosaic. It is clear that there was no transmission by the soil in which mosaic plants had just been grown, all mosaic that was shown evidently being transmitted by the tubers.

##### FIELD EXPERIMENTS

The greenhouse experiment described in the previous paragraph was not concerned with certain factors in the possible soil-harboring of mosaic in fields—namely, old stalks, volunteer potato plants, and insects. There is no doubt, when the proofs of transmission by aphids are remembered, but that volunteer mosaic plants may contribute to the infection of healthy stocks planted where mosaic stocks were grown the preceding season if they are not discovered and removed before the appearance of aphids. Even if they are, other factors might cause the infection of healthy plants.

To test this supposition, three rows of Green Mountain stock from a plot rogued in 1918 were planted across the location of a 1918 20 per cent diseased Green Mountain plot and a wholly diseased one. Each mosaic hill was dug and the seed piece examined. If volunteers are disregarded, 28 per cent of the 142 hills grown upon the ground of the all-diseased plot were mosaic as were 28 per cent of the 481 plants grown upon the ground which had produced the 20 per cent mosaic plots. This evidently was from infection the previous season, since 27 per cent of the hills were mosaic by July 15.

A similar but more extensive test consisted in planting 19 rows of the same stock across the ground which had produced 14 of the 1918 plots. Similar examination of the mosaic plants on July 30 showed 22 per cent

of the 4,466 hills to be mosaic. Although this stock was retarded in its development by being frozen nearly to the ground on June 23, only 1 per cent of the hills developed mosaic between July 30 and August 18. The nature of the various 1918 plots and the percentages of mosaic on the same ground in 1919 are given in Table VIII.

TABLE VIII.—Nature of 1918 plots and percentage of mosaic hills in the parts of the 1919 plot grown upon the same ground

Section No.	1918.		1919.	
	Variety.	Disease.	Total number of hills.	Percentage of mosaic hills from seed pieces.
1	Green Mountain.....	11 per cent mosaic.....	424	24
2	Bliss Triumph.....	15 per cent mosaic.....	432	23
3	Green Mountain.....	13 per cent mosaic.....	454	22
4	do.....	45 per cent mosaic.....	422	26
5	do.....	46 per cent mosaic.....	375	18
6	Roxbury Wilson.....	10 per cent mosaic.....	281	23
7	Bliss Triumph.....	100 per cent mosaic.....	350	23
8	Green Mountain.....	do.....	458	22
9	do.....	11 per cent mosaic.....	140	28
10	Irish Cobbler.....	No leafroll.....	143	22
11	do.....	All leafroll.....	105	22
12	do.....	No leafroll.....	169	15
13	Miscellaneous.....	Leafroll and mosaic.....	140	23
14	do.....	do.....	573	24

It will be noted that there are few marked deviations from the percentage for the whole plot, which was 23 per cent. These consist of one deviation upward and one downward for the ground occupied by two half-mosaic plots (4 and 5) and of the same for two comparatively mosaic-free plots (9 and 12) and therefore are without significance in regard to soil-harboring of the disease.

#### SUMMARY

(1) Transmission of potato mosaic by means of tubers, grafting, plant juice, and aphids was effected under various conditions, including those essentially of the field with insects controlled.

(2) Infection was obtained with intervarietal transfer of juice.

(3) Transmission was attempted, but without success so far as could be ascertained in the same season, by means of flea beetles, Colorado potato beetles, the seed-cutting knife, and contact of seed pieces, of roots, and of vines.

(4) Preliminary observations indicate that infection does not result from growth in soil that produced mosaic potato plants the previous season.

(5) It appears impossible either for infected plants to recover or, so long as diseased stock is not far off and insect carriers exist, to assure the maintenance of health of susceptible varieties by roguing plots or by selecting hills, tubers, or seed pieces.

(6) Isolation of plants by means of insect cages, as well as elimination of insects in the greenhouse, have maintained stocks disease-free, indicating that control of aphids and possibly of some other kinds of insects as well, is the most important means of checking the spread of potato mosaic among susceptible varieties.

PLATE 49

Vines of Green Mountain variety inoculated with juice from healthy foliage of the same variety. No mottling and no ruffling of leaves.

(338)





PLATE 53

Vines of Bliss Triumph variety inoculated with juice from healthy foliage of Irish  
Cobbler variety. No mosaic mottling.



PLATE 51

Vines of Irish Cobbler variety inoculated with juice from healthy foliage of the same variety. No mosaic.





PLATE 52

Vines of Green Mountain variety inoculated with juice from mosaic foliage of the same variety. Distinct mosaic mottling and ruffling of young leaves on top of stalks. For control see Plate 49.

PLATE 53

Vines of Green Mountain variety inoculated with juice from mosaic foliage of Bliss Triumph variety. Distinct mottling and ruffling of upper leaves and early dying of lower leaves. Condition of control plants same as vines in Plate 49.





PLATE 54

Vines of Green Mountain variety inoculated with juice from mosaic foliage of Irish Cobbler variety. Distinct mottling and ruffling of upper leaves, early dying of lower leaves. Condition of control plants same as vines in Plate 49.

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PLATE 55

Vines of Bliss Triumph variety inoculated with juice from mosaic foliage of Irish Cobbler variety. Mosaic mottling of upper leaves, early dying of lower leaves. For control see Plate 50.





PLATE 56

Vines of Irish Cobbler variety inoculated with juice from mosaic foliage of the same variety showing bad stage of mosaic. Distinct mottling of young leaves and early dying of foliage. For control see Plate 51.

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